PROCEEDINGS
OF THE
NATIONAL ACADEMY OF SCIENCES
Volume 26 September 15, 1940 Number 9
Copyright 1940 by the National Academy of Sciences

A NOTE ON THE VARIATION OF DELTA SCUTI

By Theodore Eugene Sterne

Harvard College Observatory

Communicated July 15, 1940

The star δ *Scuti*, $18^h36^m.8$, $-9°9'$ (1900), was found by Fath,[1,2] from photoelectric measures made during July, 1935, and July and August, 1936, to be variable with a range of the order of $0^m.2$ and a period of $0^d.19377$. The radial velocity had already been found by Colacevich[3] to be variable in that period. Neither the light curve nor the radial-velocity curve, however, is regularly periodic, and both curves appear to be in a state of continual change. From the several nights on which simultaneous light and radial-velocity measures were made, it was seen that changes in the light curve are correlated with changes in the radial-velocity curve; and, finally, Fath found that the range of the variation in light appeared to be periodic in a period of $5^d.24774$. He suggested that this might be an interference period, between the fundamental period of $0^d.19377$ and a slightly longer "secondary" period of about $0^d.20120$. Such a pair of periods would get into step with each other once every 5.24774 days, producing very nearly the observed variation in the range.

It was pointed out by Sterne[4] that the preceding periods could not reasonably be reconciled with the pulsation theory. For any single gaseous star, there are a set of dynamically possible modes of radial oscillation, which differ among themselves in the disposition and number of their nodal surfaces, and in their periods. In the mode of lowest order (mode zero), the displacement vanishes only at the center; in the next mode the displacement vanishes at the center and also over a spherical surface inside the star. The ratio of the period of mode one to that of mode zero depends upon the law of distribution of matter within the star, as well as upon the effective ratio of specific heats, γ, of the mixture of matter and radiation within the star. The ratio of periods was computed for a polytrope of index three by Edgar,[5] for certain particular values of γ. The ratio of the period of mode two to that of mode zero was computed for this same model by Miss Kluyver,[6] who adopted a value of 20/13 for γ; while Sterne,[7] in order

to determine what effect varying degrees of concentration of density would have upon the ratios of the periods, found the periods of all the modes of three other stellar models for all values of γ. The general theoretical result is that no reasonable distribution of density can yield a ratio, between the periods of the zero'th and first modes, that lies as close to unity as the ratio of $0^d.20120$ and $0^d.19377$. The order of the modes that could yield so close a ratio would have to be about the twenty-sixth or higher, and it is nearly impossible to imagine how a limited number of modes of such high order could be maintained in oscillation in a star, with the effective exclusion of modes of lower order.

To see whether it could not be reconciled with the pulsation theory, Sterne[4] rediscussed the observational material. The residuals of Fath's observations from the mean light curve, formed according to the period $0^d.19377$, were found not to be periodic in a period $0^d.20120$ and thus this value of the secondary period was ruled out. The residuals, however, could be fitted by a secondary period of $0^d.18688$, or alternatively of $0^d.15739$. A third possible value, $0^d.13603$, was not in as good agreement with the observations as either of the two longer values. The first of the three observationally plausible values was inconsistent with the pulsation theory; the last two were not obviously inconsistent with it. Fath's observations were made at the Lick Observatory, between about 0.7 and 0.9 of the Julian day, and did not enable one to discriminate between the two most observationally probable values of the secondary period. The two alternative periods fitted the observed magnitudes and the observed ranges equally well, and both fitted an observed sine-term in the ephemeris of the times of median increasing magnitude. Both values of the secondary period, moreover, seemed to fit the radial-velocity measures published by Colacevich.[3] The principal indeterminacy, which arose from the distribution of the observations in time during the hours of darkness at a single observing station, could be removed only by observations made in different longitudes. There was another, and trivial, indeterminacy arising from the seasonal distribution of the observations.

As the result of an arrangement made by Fath through correspondence with the director, δ *Scuti* was observed photoelectrically[8] by the Russian Abastumani Observatory, on Mt. Kanobili in Georgia, during the summer of 1938 while Fath observed the star at the Lick Observatory. Fath has recently discussed[9] the important Mt. Kanobili observations, along with his own. The shorter, of Sterne's pair of suggested values of the secondary period, appears to fit the combined Mt. Kanobili and Lick observations better than the longer value. The value adopted for the secondary period by Fath is $0^d.157382$. For the fundamental period he finds $0^d.193770$, and he finds evidence also for a third period, $0^d.095156$. The corresponding ranges are $0^m.033$, $0^m.167$ and $0^m.011$. Besides these three periods, that

persist throughout the three observational series, he finds small variations that do not persist throughout them all, and still others that appear to be erratic and hardly distinguishable from observational errors.

It is the purpose of this note to indicate the bearing of Fath's recent results on the pulsation hypothesis. The three observed periods are in the ratio of 1:0.812:0.491. As has been mentioned, Miss Kluyver[6] has computed the ratio of the period of the second radial mode, of a polytrope of index three, to that of the zero'th mode. The value of γ that she adopted was 1.54, and the ratio of periods that she found was 1:0.515. A slightly smaller value of γ would make the agreement with the observed ratio, 1:0.491, exact. Unfortunately, the period of the first mode of vibration of the polytrope does not appear to have been correctly computed by anyone, and a comparison with the observed ratio 1:0.812 is not now possible. The periods computed by Edgar,[5] and included with reservations by Sterne[7] in his tables, appear to be inconsistent with Miss Kluyver's computed results and to be too short.

According to the *Henry Draper Catalogue*, the visual magnitude of δ *Scuti* is 4.74 and its spectral class is *F*0. Its revised spectral class is given as *F*5 by the Lick observers and as *F*4*s* by Mount Wilson. The Mount Wilson absolute visual magnitude is +1.4. From the spectral class and absolute magnitude, the star's radius is found[4] to be about 3.6 times the sun's; and from the spectral class, absolute magnitude, and mass-luminosity-temperature relation the mass is found to be about 2.5 times the sun's. The mean density is thus about 0.076 gm./cm.^{3}, and with this mean density a polytrope of index three would have the period $0^d.19377$ for its lowest mode if γ were just about 1.54. However, the observational determination of the density is very rough, and but little stress should be laid on the close agreement between the values of γ inferred on the one hand from the ratios of periods, and on the other from the length of the fundamental period. Moreover, the polytropic model can be but an approximation to the actual distribution of density. Nevertheless, it can at least be stated that there does not now appear to be any insurmountable discrepancy between the periods of δ *Scuti* and the pulsation theory. The very small non-persistent variations (of the order of one or two hundredths of a magnitude) reported by Fath do not seem to the present author to be serious obstacles. Such small, irregular variations may not be related to pulsations at all, but may be merely superficial in nature, or due to convection currents. It seems improbable that any luminous star should be absolutely constant in brightness, and in Cepheids one should expect the effects of the important pulsations and of the small erratic disturbances to be additive.

It is greatly to be hoped that the periods of the first and second modes of a polytrope of index three may be computed for several values of γ

in order to allow a more complete comparison to be made. It is possible that some information may thus be obtained about the distribution of density, since one ratio, but not two, can be fitted by varying γ.

In view of its smallest damping,* one would in general expect the mode of longest period to exhibit the greatest amplitude. However, the theoretical problem of the relative amplitudes is complicated by the influence, upon the maintenance of the oscillations, of the manner in which the compression-sensitive sources of energy are distributed throughout the stellar interior. Sources concentrated into regions where there are no changes in temperature and density, during the oscillation of a certain mode, cannot be effective in maintaining that mode in a state of oscillation even if the rate of generation of energy increases rapidly with increasing temperature and density. On the other hand, compression-sensitive sources in regions where there are changes in temperature and density, during the oscillation of a particular mode, can be effective in maintaining that mode in oscillation. It is of some interest in this connection to notice that the ranges of the three periodic variations found by Fath in δ *Scuti* are in the ratios of 1:0.197:0.066 magnitudes, in decreasing order of periods. δ *Scuti* is a member of the β *Canis Majoris* type of stars. The existence of sine-terms, in the ephemerides of cluster-type variables like *RR Lyrae*, indicates that some of these stars also oscillate in more than one mode simultaneously. Classical Cepheids like δ *Cephei*, of still longer period, exhibit great regularity in their light curves. The differences between the numbers of modes that are oscillating in some Cepheids, and in others, doubtless arise from differences of model that may involve differences in their distributions of sources of energy and in their compositions.

Summary.—From photoelectric observations made in different longitudes, Fath has found (*Lick Observatory Bulletin*, No. 501, 1940) that the secondary period of the star δ *Scuti* is probably $0^d.157382$, and he finds evidence for a persistent third period of $0^d.095156$. It is pointed out that these periods and the fundamental period of $0^d.193770$ can be not unreasonably interpreted as the three longest periods of radial oscillation. For a more complete comparison with the pulsation theory, and to gain knowledge about the distribution of density, it is desirable for the periods of the first and second modes of a polytrope of index three to be computed for each of several values of the effective ratio of specific heats.

[1] Fath, *Lick Obs. Bull.*, **17**, 175 (1935).
[2] Fath, *Ibid.*, **18**, 77 (1937).
[3] Colacevich, *Ibid.*, **17**, 171 (1935).
[4] Sterne, *Ap. J.*, **87**, 133 (1938).
[5] Edgar, *M. N.*, **93**, 422 (1933).
[6] Kluyver, *B. A. N.*, **7**, 313 (1936).
[7] Sterne, *M. N.*, **97**, 582 (1937).

[8] Nikonov, *Bull. Abastumani Ap. Obs.*, No. 3, 27 (1938).
[9] Fath, *Lick Obs. Bull.*, **19**, 77 (1940).
[10] See, for example, reference 5.

GALACTIC AND EXTRAGALACTIC STUDIES, VIII. A NEW DETERMINATION OF THE PERIOD-LUMINOSITY CURVE

BY HARLOW SHAPLEY

HARVARD COLLEGE OBSERVATORY

Communicated August 13, 1940

1. Introduction.—The basic significance of the period-luminosity relation among Cepheid variables in all studies of the distances and dimensions of external galaxies encourages every effort to improve its empirical standardization. The coördinates of the period-luminosity curve most commonly used in recent years are those published in Harvard Monograph No. 2, p. 135 (1930). For the derivation there given, the form of the curve is based on Cepheid variables in the Small Magellanic Cloud and in the globular clusters. The zero point of the curve (which happens to give absolute magnitude zero for the median of average cluster-type variables) is based on the parallaxes and motions of the brighter galactic Cepheids. Recently the zero point has been examined critically by R. E. Wilson, who not only had at his disposal a great deal more observational data on radial velocities and proper motions than were at hand for my original derivation of the curve, but has also included in his discussion the effects of space absorption on the magnitudes and galactic rotation on the motions of galactic Cepheids.[1] Wilson finds no need as yet for altering the adopted zero point, the computed correction being 0.0 ± 0.2 for the cluster-type Cepheids and -0.14 ± 0.2 for the classical Cepheids.

TABLE 1

NEW PERIODS AND MAGNITUDES FOR THIRTEEN VARIABLES

H. V.	log *P*	$\dot{m}$	*A*	H. V.	log *P*	$\dot{m}$	*A*
1408	0.214	16.5	0.95	1943	0.210	16.75:	0.5:
1420	0.294	16.5:	>1.0	1964	0.321	16.45:	1.1:
1655	0.207:	16.75	0.4	2002	0.371	16.2	1.0
1675	0.166	16.8	0.6	2114	0.390	16.4	1.1
1731	0.327	16.5	1.0	2173	0.482	16.2	1.3
1928	0.221	16.45	0.9	3610	0.178:	16.8:	>0.6
1823	0.524	16.3	0.9				

It is now possible to reëxamine the form of the period-luminosity curve with the aid of much new material on variable stars in the two Magellanic clouds. Altogether we have available the periods and corrected median

magnitudes of 307 variables in the Small Cloud, and 137 in the Large Cloud. To those already published for the Small Cloud,[2] we can now add the list of faint variables given herewith in table 1. More than half of these thirteen stars have periods of less than two days and are valuable therefore in strengthening the fainter end of the period-luminosity curve. In fact, most of these stars had been suspected, on the basis of the insufficient observational material then at hand, to be cluster-type Cepheids, or possibly variables in the period-interval between one and two days where it had been supposed until recently that periods rarely occur.[3] Miss McKibben derived the periods and median magnitudes for the stars of table 1; she and Miss Dowse have assisted in the calculations for the present paper.

TABLE 2

INTERVAL OF LOG P	MEAN LOG P	MEAN $\dot{m}$	NO. OF STARS	WEIGHT
0.0–0.2	0.140	16.71	17	1
0.2–0.4	0.297	16.45	53	3
0.4–0.6	0.501	16.22	61	4
0.6–0.8	0.688	15.88	58	4
0.8–1.0	0.897	15.54	39	2
1.0–1.2	1.108	15.03	36	2
1.2–1.4	1.263	14.82	19	1
1.4–1.6	1.485	14.40	17	1
1.6–1.8	1.662	14.05	2	
1.8–2.0	1.877	12.98	3	
>2.0	2.212	12.85	2	

2. The Period Luminosity Curve.—The data for the individual stars are plotted in figure 1, where the vertical ordinates are absolute photographic median magnitudes and the abscissae are logarithms of the periods. Practically without exception the periods are very accurately determined, and the considerable scattering therefore represents observational and natural deviations in magnitude. The plotted crosses in the diagram represent means for intervals of two-tenths in the logarithm; table 2 contains these mean values. The fifth column of the table shows the weights adopted in the solution for the straight line in the diagram, for which the equation is

$$\dot{m} = 17.04 - 1.74 \log P \qquad (1)$$
$$\pm .05 \quad \pm .06$$

Although a straight line adequately represents the period-luminosity relation for the interval from 1.2 to 40 days, there is no *a priori* reason for expecting that the complicated phenomenon of Cepheid variation should exhibit throughout its whole range of occurrence a linear relation between $\dot{m}$ and log P. In fact, we know that the curve flattens out for periods shorter

than one day, and the present evidence of figure 1 indicates also a deviation from linearity for the longest periods.

To obtain the absolute photographic magnitude as a function of period, the zero point has been based on Wilson's work on classical Cepheids, mentioned above. From his results we find that corresponding to $\log P = 0.89$, $\dot{M} = -1.83$. From (1) we have $\dot{m} = 15.49$ for $\log P = 0.89$. On the

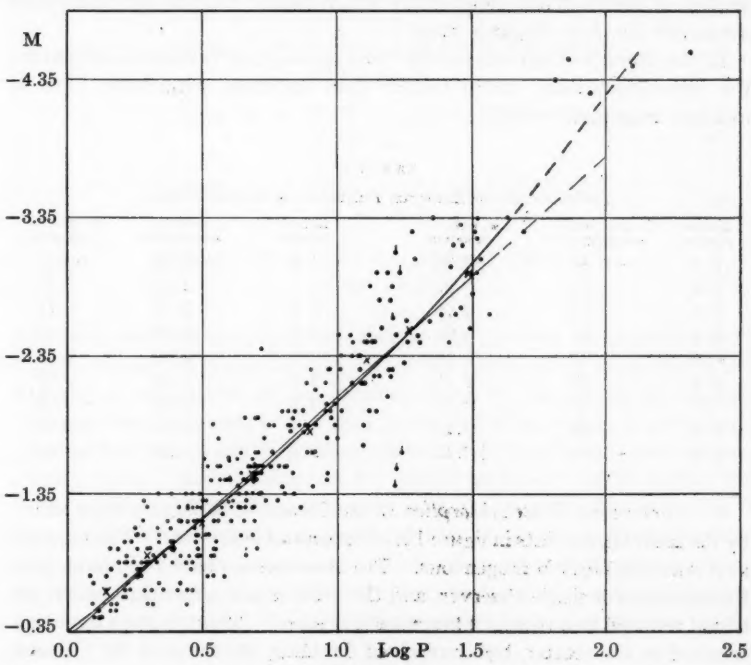

FIGURE 1

Photographic period-luminosity curve.

absolute scale, therefore, since the modulus is $\dot{m} - \dot{M} = 17.32$, the straight line representation may be written

$$\dot{M} = -0.28 - 1.74 \log P \qquad (1')$$

which will be closely approximate for periods between 1.2 and 40 days.

It seems best, however, to represent the observations with the nonlinear curve drawn in figure 1, and the coördinates of this curve are those appearing in table 3 as the best values now available for the period-luminosity relation. The last column of table 3 gives the differences between the earlier photographic period-luminosity curve and the present revision.

Not only are there few stars with periods longer than forty days, but their redness and relatively great apparent brightness make them difficult to measure with accuracy or certainty in the magnitude system applicable to the Cepheids of shorter period. The scattered points in figure 1 for log $P > 1.6$ may therefore be revised with further study of the magnitude system. A special photometric program is now under way at the Boyden Station of the Harvard Observatory which should stabilize the magnitude sequences for these brighter stars.

In the absence of periods shorter than one day, no revision is offered for the period-luminosity curve fainter than apparent magnitude 17.0, or absolute magnitude -0.35.

TABLE 3

COÖRDINATES OF ADOPTED PERIOD-LUMINOSITY CURVE

LOG OF PERIOD	ABS. PG. MAGNITUDE	H. MON. 2 −ADOPTED	LOG OF PERIOD	ABS. PG. MAGNITUDE	H. MON. 2 −ADOPTED
0.1	−0.51	+0.06	1.0	−2.02	+0.13
0.2	0.68	0.07	1.1	2.20	0.13
0.3	0.85	0.08	1.2	2.39	0.13
0.4	1.01	0.08	1.3	2.59	0.12
0.5	1.17	0.10	1.4	2.80	0.12
0.6	1.33	0.11	1.5	3.02	0.09
0.7	1.49	0.12	1.6	3.25	+0.06
0.8	1.66	0.13	1.8	3.73	−0.08
0.9	−1.84	+0.13	2.0	−4.24	−0.36

3. Concerning Space Absorption in the Cloud.—The large scatter shown by the individual points in figure 1 is of considerable interest. The material as it stands is fairly homogeneous. The observations have been reduced to the system of a single observer, and the various sequences throughout the Cloud reduced to a common magnitude system.[2] Many factors have contributed to the scatter, but unresolved doubling, thickness of the Cloud in the line of sight, space absorption in the Cloud, Eberhard and background effects and the natural spread of the luminosities for a given period are the most important. A considerable checking of results shows that the spread can be little decreased by more profuse or more precise photometry. The thickness of the Cloud can contribute about two-tenths of a magnitude to the spread. In places, local space absorption appears to play an important rôle; for example, what appears to be the effect of fairly heavy localized absorption is illustrated in figure 2 for six variables with periods almost alike, but with a spread in luminosity of more than 1.5 magnitudes. (In figure 1 these six stars are represented with dots and extruding lines.) The three faintest variables are in the denser part of the Cloud.

It might be expected from figure 2 that the median magnitudes for a given period would be systematically brighter in the outer parts of the sys-

tem because of centralized space absorption. But, after the reduction of all observations to the system of a single observer and a single magnitude sequence, no appreciable general absorption is established. To examine this matter closely, period-luminosity curves were derived separately for the inner and outer regions, and also for the nucleus and for the border regions. The resulting equations below should be compared with (1), and with each other.

$$\text{Inner:} \quad \dot{m} = \underset{\pm.06}{17.00} - \underset{\pm.07}{1.69} \log P \ (143 \text{ stars}) \tag{2}$$

$$\text{Outer:} \quad \dot{m} = \underset{\pm.09}{17.06} - \underset{\pm.12}{1.79} \log P \ (164 \text{ stars}) \tag{3}$$

$$\text{Nuclear:} \quad \dot{m} = \underset{\pm.12}{16.84} - \underset{\pm.11}{1.44} \log P \ (77 \text{ stars}) \tag{4}$$

$$\text{Border:} \quad \dot{m} = \underset{\pm.08}{17.01} - \underset{\pm.10}{1.70} \log P \ (80 \text{ stars}) \tag{5}$$

The mean errors of the parameters indicate the fit of the calculated straight lines to the normal points for intervals of two-tenths of the logarithm of the period. A comparison of the constant terms in the foregoing relations indicates that the absorption in the nucleus and inner regions is not greater than farther out; if anything, the nucleus is the clearest—a somewhat surprising result. But the slope of the period-luminosity curve is also low in the nucleus. Notwithstanding the large mean error, this low value is probably significant, and is indirectly attributable perhaps to the remarkable period-frequency phenomenon discussed in an earlier paper:[3] among the nuclear stars there are relatively few short-period classical Cepheids. If we take a mean slope of 1.57 for the nuclear and border regions, we obtain:

$$\text{Nuclear:} \quad \dot{m} = \underset{\pm.05}{16.95} - 1.57 \log P$$

$$\text{Border:} \quad \dot{m} = \underset{\pm.05}{16.94} - 1.57 \log P$$

An extended study of the variables in nuclear regions is now in progress, with the hope of throwing more light on equation (4), as well as on the frequency of periods and on internal absorption.

Another test of the possible differences in space absorption, when inner and outer regions are compared, is obtained directly from the mean apparent magnitudes of the variables with periods between five and ten days. We have:

	MEAN PERIOD	MEAN MAGNITUDE	NUMBER OF STARS	REDUCED MAGNITUDE
Inner	7.130	15.60	41	15.60
Outer	6.899	15.68	20	15.66
Nuclear	7.126	15.65	27	15.65
Border	6.785	15.75	11	15.72

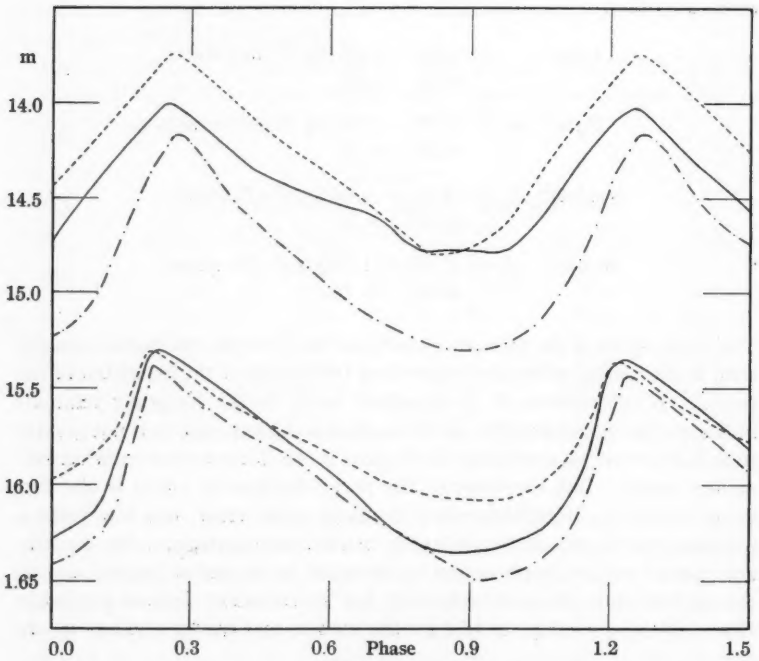

FIGURE 2

Illustration of local space absorption in Small Magellanic Cloud. The variables in descending order (left-hand margin) are H. V. 1954, 1925, 1787, 1478, 1533 and 828. The periods, in days, are respectively, 16.7, 16.8, 16.2, 17.5, 16.4, 16.3. Minimum magnitude for H. V. 1478 is considerably brightened by a partially unresolved companion star. H. V. 1925, also of small range, is not visibly double. The three faintest variables are in the densest part of the nucleus; the three brighter variables are well out of the dense nucleus, but not in border areas.

where the last column contains magnitudes reduced to the period 7.13 days. The result is the same as before, since, in photographic magnitudes,

$$\text{Inner} - \text{Outer} = -0.06 \pm 0.06 \text{ (m. e.)}$$

$$\text{Nuclear} - \text{Border} = -0.07 \pm 0.06 \text{ (m. e.)}$$

and the center of the Cloud is if anything the clearest part, so far as these mean results are an adequate test of absorption.

For the 137 stars for which periods have been determined in the Large Magellanic Cloud[1] the linear relation is

$$\dot{m} = \underset{\pm.08}{17.14} - \underset{\pm.09}{2.08} \log P \qquad (6)$$

The higher slope of the curve may indicate merely that the magnitude system differs from that of the Small Cloud, notwithstanding our earlier attempts to harmonize the magnitude sequences. The new southern photometry should clear up this discrepancy.

4. Distance of the Small Cloud.—With the revised period-luminosity relation we are able to make a revised determination of the distance of the Small Magellanic Cloud. The new modulus is

$$\dot{m} - \dot{M} = 17.35,$$

a value differing inappreciably from 17.32, derived ten years ago. Although the modulus is well determined, with a mean error, probably not exceeding a tenth of a magnitude, the distance of the Cloud is indeterminate over a considerable range because of uncertainties as to the correction necessary to allow for galactic space absorption. The galactic latitude of 45° insures fairly high transparency; and from available long-exposure Bruce photographs we have evidence of a high population of external galaxies in the neighborhood of both Magellanic Clouds.[4] But much further analysis of stellar and nebular distribution in the southern hemisphere will be necessary before the distances to the Clouds can be given with close accuracy. For the present we may compute the distance of the Small Cloud on three assumptions as to the total photographic absorption, suggesting that the middle value be used for the present:

ABSORPTION, m	DISTANCE IN KILOPARSECS
0.0	29.5
0.25	26.3
0.5	23.4

5. Summary.—The number of classical Cepheids in the Small Magellanic Cloud for which we know the periods and median magnitudes has been much increased recently, and with the standardization of magnitude sequences throughout the Cloud it has been possible to revise the period-luminosity curve. Recent work by Dr. R. E. Wilson on the zero point of the period-luminosity curve makes the revision especially appropriate at this time.

Throughout the interval from 1.2 to 40 days (which covers the periods of most of the variables known in the Magellanic Clouds), a linear relation

between the logarithm of the period and the median magnitude is fairly satisfactory. A non-linear curve is adopted (table 3) which differs but little from the one now in general use. A study of the period-luminosity relation throughout the Small Cloud reveals no measurable difference depending on the density of star distribution.

The scattering of points about the adopted curve (Fig. 1) is significant, and only a part can be attributed to "Cloud thickness." Possibly space absorption within the Cloud is spotty in distribution and accounts for some of the larger deviations, but there seems to be a real dispersion in absolute luminosity for Cepheids of the same period (Fig. 2). The new value obtained for the distance of the Small Cloud (section 4) is affected by the uncertain information on space absorption in the galactic system.

[1] Mt. Wilson Contr. No. 604 (1939).

[2] Shapley and McKibben, *Harv. Circ.* 439, 1–5 (1940).

[3] See fifth paper of this series, these PROCEEDINGS, **26,** 105 (1940); *Harvard Reprint* 192.

[4] Shapley, *Harv. Ann.*, **105,** No. 8 (1937).

---

*THE SPECTRUM OF BD +30°3639**

BY P. SWINGS AND O. STRUVE

McDONALD OBSERVATORY, UNIVERSITY OF TEXAS, AND YERKES OBSERVATORY, UNIVERSITY OF CHICAGO

Communicated August 9, 1940

Recent investigations have again directed the attention of astronomers to this remarkable object consisting of a carbon Wolf-Rayet nucleus of type *WC*8, which is surrounded by a nebula showing strong forbidden lines of ionized nitrogen. O. C. Wilson[1] has used Wright's measurements of the nuclear bands in his discussion of the physical characteristics of the Wolf-Rayet stars. Wright had called attention to the red shift of some of the bands and had attributed it to the encroachment of the absorption components on the violet edges. Wilson has also advanced an alternative explanation based upon the interpretation of the red shift as a gravitational effect, and has stressed the importance of wave-length measurements in this object.

We have recently found[2] a second object, HD 167362, which, like BD +30°3639, shows the rather peculiar association of an exciting Wolf-Rayet nucleus without any trace of nitrogen with a surrounding nebulosity which is very rich in nitrogen. This star has also fairly narrow emission bands. We secured in September, 1939, several slit spectrograms of Campbell's ob-

TABLE 1

EMISSION LINES DUE TO THE NUCLEUS OF BD 30°3639

λ	INT.	IDENTIFICATION ELEMENT	IDENTIFICATION λ	IDENTIFICATION INT.	VIOLET EDGE	RED EDGE	λ WRIGHT	λ PLASKETT	NOTES
3332.8	2	O III	3333.00	4					
		O III	3330.40	4					
3340.1	3	O III	3340.74	6			3342.		
3347.9	1	O IV	3348.08	2					
		O IV	3349.11	3					
3359.4	1	.....	.....	..					
3375.5	1	O IV	3375.5	2					
3380.2	1	O IV	3381.3	4					
3385.7	1	O IV	3385.6	5					
		O III	3384.95	4					
3391.0	2	O II	3390.25	8					
3396.6	2	O IV	3396.8	2					
3404.1	3	O IV	3403.58	3			3404.		
3412.9	4	O IV	3411.76	4			3414.		
3429.1	2	O III	3428.67	3			3429.		
		O III	3430.60	4					
3445.4	3*nn*	O III	3440.39	4	3439.6	3452.0	3447.		
		O III	3444.10	5					
		O III	3446.73	2					
		O III	3450.94	4					
3456.	1	O III	3455.12	5					
3470.	1	O II	3470.81	8					
3503.7	2*nn*	.....	.....	..					
3554.	2	He I	3554.5	1					(1)
3561.	2	O IV	3560.42	1					(1)
		O IV	3563.36	2			3564.		
3567.	2	.....	.....	..					(1)
3586.8	1	He I	3587.4	1					
3609.42	5	C III	3609.61	5	3606.1	3612.2	3611.		
		C III	3608.96	4					
3640.0	2*n*	O III	3638.70	3			3640.		
3665.6	2	.....	.....	..					
3703.91	3*nn*	O III	3702.75	5			3705.		(2)
		O III	3703.37	5					
		O III	3704.73	3					
		O III	3707.24	6					
		C III	3703.52	2					
		He I	3705.00	3					
3712.29	2	O III	3712.48	2	3711.5	3716.4			
3714.53	3	O III	3715.08	6					
3760.13	5	O III	3759.87	9	3757.0	3763.5	3760.		
3774.02	2	O III	3774.00	6	3772.5	3778.5			
3790.95	2	O III	3791.26	6	3788.4	3793.7	3794.		
3804.14	2	.....	.....	..					
3813.0	1	He II	3813.53						(3)
		O III	3810.96	2					

TABLE 1 (*Continued*)

EMISSION LINES DUE TO THE NUCLEUS OF BD 30°3639

λ	INT.	IDENTIFICATION ELEMENT	IDENTIFICATION λ	IDENTIFICATION INT.	VIOLET EDGE	RED EDGE	λ WRIGHT	λ PLASKETT	NOTES
3817.5	1	O III	3816.75	1					(3)
		He I	3819.61	4					
3888.77	5	He I	3888.65	10	3883.2	3894.3	3884*E*		(4, 5)
		C III	3889.18	4			3894*E*		
3916.20	4	.....	.....	..					
3919.81	4	C II	3918.98	6			3915*E*		
		C II	3920.68	8	3913.0	3927.2	3920*M*	3920.2	
3924.22	2	Si III	3924.44	4			3925*E*		
		He II	3923.51						
3934.53	1–2	C IV	3934.1						
3951.39	1	?O III	3951.82	0					
3961.66	2	O III	3961.59	8			3962.		
3974.30	1	O II	3973.27	10					
4026.13	3	He I	4026.19	5			4027.	4026.	(3, 5)
		He II	4025.64		4023.2	4035.6			
4031.61	2	.....	.....	..					
4056.58	3	C III	4056.06	5	4052.3	4060.6	4056.7	4056.5	(5)
4070.59	4*n*	C III	4067.87	9	4065.0	4075.2	4070.	4068.	(3, 4, 5)
		C III	4068.97	10				4071.	
		C III	4070.30	10					
4088.49	2	Si IV	4088.86	10	4085.2	4093.1	4088.9	4089.	(5)
4115.04	3	Si IV	4116.10	8			4113*E*		
							4115.1*M*	4117.	
4120.41	3	He I	4120.81	3			4121.4*M*		
		C III	4122.05	3	4113.2	4133.6			
4128.41	1–2	Si II	4128.05	8			4129.6*M*	4127.	
4130.74	2	Si II	4130.88	10			4133. *E*	4130.	
4142.85	1–2	He I	4143.77	2					
4155.94	4	C III	4156.50	4	4152.1	4158.9	4155.4	4155.	
		C III	4152.43	3					
4162.90	3	C III	4162.80	5	4159.7	4166.1	4163.8	4164.	
4168.56	1	He I	4168.97	1					
4176.99	1	.....	.....	..					
4186.44	5	C III	4187.15	10	4182.3	4191.3	4187.7	4186.	(4)
4200.90	2	He II	4199.87	2			4200.5	4200.2	
4213.	1	Si IV	4212.44	3					(6)
4228.64	2	.....	.....	..			4229.6	4229.	
4237.77	1–2	.....	.....	..			4239.		
4253.08	1	O II	4253.98	8*n*					(6)
4267.60	5	C II	4267.27	10	4260.2	4273.8	4266.9	4267.2	
		C II	4267.02	8					
4316.16	2	O II	4317.16	8			4316.7	4317.	
		C II	4317.42	4					
4325.23	5	C III	4325.70	8	4321.2	4329.9	4325.8	4325.	(5)
4347.40	2	O II	4345.57	7			4347.	4349.	
		O II	4347.43	5					
		O II	4349.43	8					

TABLE 1 (*Continued*)

EMISSION LINES DUE TO THE NUCLEUS OF BD 30°3639

λ	INT.	IDENTIFICATION ELEMENT	IDENTIFICATION λ	IDENTIFICATION INT.	VIOLET EDGE	RED EDGE	λ WRIGHT	λ PLASKETT	NOTES
4367.68	–4	C III	4368.14	4*d*			4368.1	4368.	
4376.16	1	O III	4376.15	1*n*					
		C II	4374.28	5				4379.	
		C II	4376.78	2*d*					
4382.05	3	.....	.....	..					(3)
4387.69	3	He I	4387.93	3			4385(*nn*)	4388.1	(3)
		C III	4388.24	2					
4413.95	3	C II	4411.52	5				4413.9	(3)
		C II	4411.20	5			4416.2		
		O II	4414.89	10					
4419.95	2	.....	.....	..				4417.6	
4441.20	4	C IV	4441.81	0			4441.4	4440.5	(7)
4457.21	1*nn*	.....	.....	..			4457.	4454.	(8)
								4459.	
4472.27	3	He I	4471.48	6			4472.4	4471.4	(4, 5)
4516.12	4	C III	4516.93	4			4516.6	4516.4	(4)
		C III	4516.02	3					
4542.03	2–3	He II	4541.63	5			4542.7	4542.2	(5)
4553.0	1	Si III	4552.61	9			4555.0	4554.1	
4568.0	1	Si III	4567.83	7			4569.	4567.8	
4593.06	2*n*	C III	4593.47	2*d*			4593.9		
		O II	4590.98	9					
		O II	4596.19	8					
4619.22	2*n*	C II	4618.85	5*d*			4619.	4620.	
4631.65	2	Si IV	4631.38	3*n*			4634.	4633.7	
4650.20	10	C III	4647.40	20	4645.0	4654.1	4651.6	4650.6	(4, 5)
		C III	4650.16	19					
		C III	4651.35	18					
4655.33	4	Si IV	4654.14	4*n*				4656.4	(3)
		C IV	4658.64	5					
		C IV	4656.5						
4665.74	4	C III	4665.90	6			4666.0	4665.	(4)
		C III	4663.53	4					
		C IV	4664.5						
4686.57	4	He II	4685.81	300			4687.1	4686.3	(4, 5)
4704.	1–2	.....	.....	..			4702.2		(5)
4785.7	1	C IV	4785.6				4786.4	4785.5	
		O IV	4783.4	2					

Notes:

E = edge; *M* = maximum.

(1) The separation of these three lines is difficult.

(2) Very broad band with complex structure; complicated also by the presence of a line of nebular origin.

(3) Measurement difficult.

(4) Absorption components measured at: 3882.16 (5); 4063.06 (2); 4180.93 (2); 4465.19 (2); 4510.27 (2); 4641.74 (7, from 4637.82 to 4645.03); 4660.28 (2); 4680.04 (2).

(5) Absorption components measured by Wright at: 3883.0, 4022, 4050.9, 4063.8, 4084.7, 4320.6, 4465.8, 4538, 4641.8, 4681.0, 4695.7.

(6) Line uncertain.

(7) This line is narrower than the C III lines and has a weak violet absorption component.

(8) This very broad line may be double.

---

ject at the McDonald Observatory. The nuclear lines of BD +30°3639 are relatively narrow and the surrounding nebulosity is not too rich in lines. The star is, therefore, suitable for accurate measurements of wave-lengths. One of the spectrograms was obtained with the 500-mm. camera and the quartz prisms, the dispersion being 40 A/mm. at λ 3933; another quartz spectrogram was taken with a dispersion of 100 A/mm. at λ 3933.

The spectrum has been investigated by Wright,[3] Plaskett,[4] Beals,[5] Edlén[6] and Stoy.[7] Our spectrograms are intended to supplement the earlier material. The sharp lines of nebular origin have been used to correct the radial velocity.[8]

Table 1 contains the results of measurements of the emission lines due to the Wolf-Rayet nucleus. It is limited to the region from λ 3330 to λ 4786; for the region λ <3330, we refer to the investigation by Stoy. Above λ 4786, our measurements are in good agreement with Wright's table and are, therefore, not reproduced. In the region covered by table 1, Stoy has measured lines at λλ 3342, 3358, 3375 and 3385.

The first column of table 1 gives the measured wave-lengths with two decimals when obtained from the spectrogram of higher dispersion; the ultra-violet lines given with one decimal or with no decimal were measured on the spectrogram of low dispersion. Columns 6 and 7 give the edges of some of the lines. Columns 8 and 9 contain the wave-lengths by Wright[3] and by Plaskett.[4] Most of the lines are satisfactorily identified.

Table 1 shows that the nucleus of Campbell's object is a typical carbon star; there is no trace of nitrogen at any stage of ionization.

For the discussion of a possible red shift, the only suitable nuclear features are the strong unblended lines, for which we have good identifications and reliable laboratory wave-lengths and which are not too close to lines of nebular origin. Only the lines measured with the higher dispersion are here considered. About fifteen lines may be used for this purpose. It is obvious that there is no systematic red shift. An appreciable displacement toward the red is observed only in the few cases where the violet absorption component is very strong.

We have also considered our results of measurements of two other planetary nuclei with narrow bands, namely NGC 6543 and HD 167362.

The first Wolf-Rayet nucleus contains N IV and C IV with similar intensities, whereas the second is a pure carbon star. In neither case is there an appreciable systematic red shift.

If we consider the best unblended identifications, we find the following average widths:

TABLE 2

WIDTHS OF THE BRIGHT LINES OF VARIOUS ELEMENTS

ELEMENT	IONIZATION POTENTIAL	WIDTH IN KM./SEC.
C^+	24.26	955
He	24.46	859
Si^{+++}	44.95	577
C^{++}	47.64	566
O^{++}	54.62	474

This table illustrates the well-known decrease in width with increasing ionization potential. The lines do not show an occultation effect, so that the sequence in width corresponds to a sequence in ejection velocity.

A direct determination of the ejection velocity may be made for He I and C III, for which absorption components of the P Cygni type have been observed. The result is: for He I, $V_{ej.} = 488$ km./sec.; for C III, $V_{ej.} = 378$ km./sec. This confirms the relation with ionization potential.

* Campbell's hydrogen envelope star, HD 184738; α (1900) $19^h30^m.8$; δ (1900) $+30°18'$.

[1] *Ap. Jour.*, **91**, 394 (1940).

[2] *Proc. Nat. Acad. Sci.*, **26**, 454 (1940).

[3] *Pub. Lick Obs.*, **13**, 220 (1918).

[4] *Pub. Dom. Ap. Obs.*, Victoria, **2**, 287 (1924) (especially table 18, p. 348).

[5] *Ibid.*, **4**, 271 (1929).

[6] *Zs. f. Ap.*, **7**, 378 (1933) (see table 9).

[7] *Pub. Astr. Soc. Pac.*, **47**, 162 (1935).

[8] The sharp-line spectrum consists of the usual lines of a low-excitation nebula; the Balmer series is seen to H_{22} on our low-dispersion spectrogram.

GALACTIC AND EXTRAGALACTIC STUDIES, IX. A PHOTOMETRIC SURVEY OF 22,000 GALAXIES IN THE ZONE FROM $\delta = +41°$ TO $\delta = +46°$

By Harlow Shapley and Rebecca B. Jones

Harvard College Observatory

Communicated August 13, 1940

1. Introduction.—In an earlier paper a report was made on the space distribution of galaxies in high southern galactic latitude.[1] Radial and transverse density gradients were considered, and an average space-density parameter was derived. The five hundred square degrees covered in that photometric survey are sufficiently near the south galactic pole to leave us essentially free from the difficulties of space absorption.

In the present study we examine the distribution, to photographic magnitude 17.5, and fainter, of the galaxies in a belt which encircles the sky at declination $+43^{\circ}.6$, and is therefore principally in the northern galactic hemisphere. A zone in this particular declination is selected because the plates can be made when the fields are near the Oak Ridge zenith and because the Selected Area sequences,[2] an hour apart in declination $+45°$, are available for magnitude work. More than half of the plates contain these standard magnitude sequences, well located for direct use in estimating nebular magnitudes, or for comparison (in galactic latitudes higher than 20°) with the sequences set up by the star-count method. This direct tie-up all around the zone with a consistent set of standards provides what is perhaps the most homogeneous collection of nebular magnitudes yet obtained.

2. The Observations.—The forty-four three-hour photographs used for the survey were made by Mr. Henry A. Sawyer with the 16-inch Metcalf telescope at Oak Ridge during the past five years. The marking of new galaxies and the estimates of photographic magnitudes were nearly all made by Miss Rebecca Jones. The plate material is described in table 1. It will be noted that the belt crosses the galactic plane twice, and that the highest northern galactic latitude attained by the plate centers is $+74^{\circ}.0$, the highest southern, $-18^{\circ}.2$. The material is therefore suitable for examining the galactic latitude effect (space absorption), while still keeping clear of the numerous clusters near the north galactic pole.

In form, table 1 closely follows the first table of Harvard Reprint 194.[1] The coördinates, equatorial and galactic, of the plate centers are for the equinox 1900; the qualities (Q) in the sixth column are on the scale of 10 for the highest quality; m_s and m_n are, respectively, the plate limit for stars and the limit of nebular completeness, and $N_{15.5}$ is the total number of objects equal to and brighter than magnitude 15.5.

TABLE 1

SUMMARY OF PLATES, MAGNITUDE LIMITS AND COUNTS

PLATE	RA (1900)	DEC.	λ	β	Q	m_g	m_m	N_{tot}	$N_{18.5}$	N_{25}
MC 27466	$0^h31^m.5$	+43°56′	88°.4	−18°.0	6	18.3	17.6	200	5	159
27463	1 01.2	44 04	94.0	−17.8	5	18.2	17.7	301	18	240
29210	1 36.2	43 07	100.7	−17.7	7	18.2	17.5	224	11	163
29232*	2 10.6	43 20	106.9	−15.7	5	18.2	17.5	363	39	275
27881	2 41.1	43 51	111.9	−13.0	7	18.2	17.5	442	30	356
30616*	3 13.0	43 38	117.1	−10.2	7	18.0	17.5	305	59	225
27899†	3 48.1	43 59	122.0	− 6.2	6	17.8		40	1	27
29214	4 15.7	43 13	126.2	− 3.3	6	17.8		25	0	13
29211	4 49.0	43 19	130.1	+ 1.3	5	18.0		12	1	7
27586	5 20.2	43 16	133.5	+ 5.9	4	18.0		16	0	13
27909	5 57.3	44 15	136.1	+12.1	5	18.2		29	2	20
27546	6 30.4	44 01	138.8	+17.4	7	18.6	17.5	185	18	139
28733	7 05.0	44 00	141.1	+23.3	6	18.3	17.7	570	7	468
28753	7 36.8	44 01	142.7	+28.8	6	18.2	17.5	571	10	453
29493	8 15.0	43 39	144.5	+35.5	6	18.0	17.6	711	21	583
29215	8 38.5	44 01	144.4	+39.8	6	18.2	17.7	926	36	716
27687	9 17.9	43 23	145.2	+46.9	8	18.4	17.7	835	35	663
30078	9 47.6	43 47	143.6	+52.2	8	18.5	17.9	1226	41	1028
29265	10 22.1	43 29	141.5	+58.3	6	18.4	17.5	1377	32	1096
29283	10 54.8	43 27	136.8	+63.8	8	18.5	17.5	1016	35	811
28788	11 25.2	43 43	128.7	+68.3	7	18.1	17.5	769	16	594
27639	11 58.1	43 47	115.3	+72.1	9	18.3	17.4	1277	49	974
27704	12 27.6	43 45	98.2	+74.0	5	18.1	17.5	878	43	677
30038	13 04.1	43 42	74.4	+73.6	7	18.0	17.5	1435	31	1172
30129	13 41.8	43 26	55.2	+70.2	6	18.1	17.5	1079	38	890
29962	14 13.4	43 59	46.7	+65.4	6	18.3	17.5	714	19	583
29979	14 42.8	43 51	41.0	+60.8	6	18.1	17.4	639	59	474
28949	15 10.5	43 25	37.2	+56.2	6	18.0	17.3	550	67	415
30079	15 48.9	44 00	36.0	+49.2	7	18.2	17.6	1726	69	1425
30186*	16 25.3	43 34	34.8	+42.7	6	18.0	17.5	1143	50	939
29537	16 54.4	43 48	35.5	+37.5	6	17.9	17.2	500	49	367
30123*	17 26.6	43 29	36.0	+31.6	8	18.3	17.5	808	40	616
30224	17 58.1	43 32	37.5	+26.1	6	17.7	17.2	442	14	373
28950	18 33.9	43 08	39.2	+19.8	6	18.0		222	24	189
30086	19 05.3	43 45	42.1	+14.7	7	17.9		282	23	243
30229	19 38.9	43 27	44.7	+ 9.2	7	17.9		83	10	69
30210	20 10.3	43 29	47.8	+ 4.4	8	17.9		11	1	7
29673	20 46.8	43 22	51.8	− 0.9	5	17.9		8	0	4
29670	21 14.9	43 31	55.4	− 4.4	6	17.9		11	0	9
27412	21 48.4	43 18	59.9	− 8.6	7	18.3		61	5	42
29661	22 19.8	43 02	64.6	−12.0	6	17.9		41	13	30
27880	22 51.3	43 47	70.1	−14.0	6	18.2		59	4	44
29760	23 27.1	43 30	76.3	−16.6	8	18.0	17.4	148	17	108
29768	23 58.0	43 04	81.9	−18.2	6	18.3		51	5	36
					6.4	18.12	17.51	22,311	1047	17,735

* Contains recognized cluster of galaxies.

† Exposure 155 minutes; all others 180 minutes.

TABLE 2

SUMMARY OF MAGNITUDES

PLATE	]15.5	15.5	15.6	15.7	15.8	15.9	16.0	16.1	16.2	16.3	16.4	16.5
MC 27466	2	3	5	5	10	15	20	25	33	35	36	38
27463	13	14	16	17	22	23	25	27	29	32	36	41
29210	9	10	11	13	15	15	15	16	22	26	30	35
29232	27	31	33	41	46	51	54	59	69	77	85	93
27881	19	23	29	31	37	43	51	59	68	78	85	93
30616	30	38	45	61	67	73	77	79	83	93	102	113
27899	1	1	1	2	6	7	8	10	12	12	13	15
29214	0	0	0	0	0	0	0	0	0	0	0	0
29211	0	1	1	2	2	3	3	4	4	4	4	4
27586	0	0	1	1	2	3	3	4	4	6	6	7
27909	2	2	2	2	2	3	3	4	6	6	9	14
27546	11	13	15	17	21	23	27	31	36	41	44	48
28733	4	4	6	8	11	16	20	24	34	48	59	69
28753	5	6	8	14	21	28	36	46	59	73	87	104
29493	15	16	18	23	32	39	48	56	64	78	89	102
29215	27	33	42	50	55	59	69	80	90	104	116	128
27687	27	29	34	44	56	68	79	90	107	121	134	149
30078	29	32	34	42	55	66	77	87	101	119	136	156
29265	25	27	32	43	59	66	74	82	92	106	121	137
29283	22	29	39	41	49	56	66	77	93	116	137	159
28788	8	12	16	19	26	34	43	52	60	74	86	99
27639	43	49	55	60	70	75	88	99	109	121	140	188
27704	25	30	32	39	45	57	69	81	95	115	129	142
30038	21	25	35	45	55	64	77	93	120	154	177	197
30129	26	28	34	36	44	53	61	69	76	98	113	128
29962	14	16	20	23	29	36	42	49	60	74	89	107
29979	38	42	44	51	56	64	71	77	87	101	111	120
28949	45	51	60	63	68	79	93	107	121	132	148	167
30079	39	48	58	71	86	107	128	146	164	182	200	222
30186	39	46	53	68	91	109	125	141	167	199	225	253
29537	24	31	42	58	71	80	96	112	129	150	164	177
30123	20	25	29	33	45	53	64	76	90	106	123	142
30224	9	13	17	27	40	50	64	80	105	121	138	158
28950	12	18	26	33	38	45	53	62	74	84	94	107
30086	18	21	24	27	32	38	46	56	69	80	94	110
30229	7	9	11	14	17	18	21	26	33	37	42	47
30210	1	1	1	2	2	2	2	3	3	3	3	3
29673	0	0	0	0	0	1	2	3	3	3	3	3
29670	0	0	1	1	1	1	2	3	4	5	6	8
27412	1	1	1	1	4	7	10	13	19	23	25	26
29661	10	11	13	13	14	17	19	21	23	24	24	25
27880	1	2	2	2	2	2	3	5	6	12	15	19
29760	12	14	16	18	22	24	26	29	31	32	34	38
29768	2	3	5	10	12	16	18	18	19	22	24	25
Cum. tot. 44	683	808	967	1171	1438	1689	1978	2281	2673	3127	3536	4016
Log tot. 44	2.83	2.91	2.99	3.07	3.16	3.23	3.30	3.36	3.43	3.50	3.55	3.60
Log tot. 18	2.69	2.76	2.83	2.91	3.00	3.07	3.14	3.21	3.26	3.33	3.39	3.44
Log tot. 9	2.35	2.41	2.49	2.55	2.64	2.70	2.77	2.83	2.90	2.98	3.04	3.11
Non-cum. tot.	683	125	159	204	267	251	289	303	392	454	409	480

16.6	16.7	16.8	16.9	17.0	17.1	17.2	17.3	17.4	17.5	17.6	17.7	17.8	[17.8
43	46	50	60	68	78	84	93	101	117	133	143	152	159
45	49	55	64	71	77	91	105	122	150	171	198	224	240
45	50	55	64	70	76	87	93	102	120	138	149	153	163
104	111	121	130	136	141	156	166	185	213	238	251	266	275
121	135	152	170	189	214	239	253	271	294	315	333	344	356
123	128	135	142	147	153	161	171	185	201	213	219	223	225
16	16	17	20	22	24	27	27	27	27	27	27	27	27
0	0	1	2	3	4	7	9	11	12	13	13	13	13
4	4	4	5	5	6	6	6	6	6	7	7	7	7
8	8	9	9	10	12	12	13	13	13	13	13	13	13
16	17	18	19	19	20	20	20	20	20	20	20	20	20
61	66	72	78	82	88	94	104	110	118	125	130	134	139
82	94	110	128	146	170	203	225	254	291	335	380	426	468
124	140	160	180	196	213	248	272	304	356	391	421	443	453
117	130	146	168	188	213	251	300	352	435	502	561	581	583
141	154	171	191	211	238	284	315	364	431	504	597	664	716
182	205	233	247	263	288	316	341	370	422	472	531	580	663
192	209	228	264	293	325	376	433	486	567	647	732	831	1028
164	177	192	228	268	323	391	470	573	707	808	886	956	1096
185	203	225	264	297	338	403	461	523	607	664	715	758	811
122	140	162	189	214	243	285	332	386	452	508	542	571	594
225	254	291	324	401	467	537	610	701	778	860	919	956	974
176	192	210	233	259	297	336	386	437	495	561	614	653	677
245	272	304	368	426	495	586	679	789	933	1024	1112	1157	1172
158	177	202	236	270	316	388	457	546	637	704	763	825	890
124	145	174	212	239	265	319	360	409	460	499	534	560	583
146	161	179	202	225	254	293	318	354	395	431	457	469	474
186	197	211	233	252	274	312	337	358	387	401	411	414	415
248	269	295	345	392	453	571	685	800	942	1096	1246	1352	1425
294	319	348	391	426	466	532	596	665	774	847	903	933	939
201	213	226	248	266	286	319	338	354	363	365	367	367	367
168	184	204	230	251	274	320	356	402	460	502	554	589	616
183	203	229	259	280	299	331	349	364	371	373	373	373	373
123	134	147	154	162	175	178	183	187	188	188	188	188	189
134	151	172	193	208	223	237	239	243	243	243	243	243	243
53	56	60	63	65	66	69	69	69	69	69	69	69	69
5	5	6	6	6	6	6	7	7	7	7	7	7	7
3	3	3	3	3	3	3	3	4	4	4	4	4	4
8	8	9	9	9	9	9	9	9	9	9	9	9	9
30	32	34	37	39	40	42	42	42	42	42	42	42	42
27	27	28	29	29	30	30	30	30	30	30	30	30	30
22	24	28	32	35	38	39	43	44	44	44	44	44	44
44	48	54	60	65	71	76	86	96	104	106	107	107	108
29	30	32	32	32	32	33	36	36	36	36	36	36	36
4727	5186	5762	6521	7238	8083	9307	10427	11711	13330	14685	15900	16813	17735
3.67	3.71	3.76	3.81	3.86	3.91	3.97	4.02	4.07	4.12	4.17	4.20	4.23	4.25
3.52	3.56	3.60	3.66	3.71	3.76	3.83	3.89	3.95	4.01	4.06	4.09	4.12	4.15
3.19	3.24	3.29	3.35	3.41	3.48	3.55	3.61	3.67	3.74	3.78	3.82	3.84	3.86
711	459	576	759	717	845	1224	1120	1284	1619	1355	1215	913	922

Table 2, in which detailed photometric results are summarized, is analogous to the corresponding tabulation in the earlier paper, except that cumulative totals are given in place of numbers at each tenth of a magnitude. The lines at the bottom of the table give the magnitude frequency (as cumulative totals) directly for all 44 plates, and logarithmically for these 44 as well as for 18 and 9 plates of highest latitude. The last line gives the non-cumulative totals for all the plates.

3. Discussion.—An 8 × 10-inch plate made with the Metcalf refractor covers fairly successfully thirty-five square degrees; but, in an analysis of the distribution of galaxies, only the central twenty-five square degrees are commonly used, in order to avoid the uncertain corrections for distance from the plate center that would be necessary if the eastern and western edges of the plate were also included. The loss at these edges can be de-

					MEAN						MEAN
362	340	320	324	279	325	258	251	241	228	173	230
278	271	289	281	269	278	204	208	207	214	193	205
281	257	250	288	284	272	171	176	184	225	208	193
290	276	298	240	302	281	206	189	206	176	212	198
301	312	303	302	297	303	231	230	224	221	222	226
302	291	292	287	286	292	MEAN	214	211	212	213	202	210

FIGURE 1

Total numbers of galaxies brighter than magnitude 17.1 in each square degree for 44 plates (left), and for 18 plates of highest latitude.

duced from the totals in table 1. Thus for all the plates together there are 16.1 galaxies per square degree for the central twenty-five square degrees, and only 10.4 galaxies per square degree for the strips at the east and west ends of the plate. This difference corresponds, on the average, to an effective loss of nearly one-third of a magnitude; it is partly true magnitude loss, partly failure to discover faint galaxies in an area of slightly distorted images.

To examine possible distance-correction effects within the area of twenty-five square degrees we have constructed figure 1, wherein the results of all 44 plates have been collected in a single checkerboard array, and also separately for the 18 plates with highest galactic latitudes. North is at the top, east on the left. These arrays include, for each of the twenty-five squares, the number of galaxies for which the mean of the twice-estimated magni-

tudes is 17.0 and brighter. Before assembling the arrays, appropriate corrections were made to eliminate from the counts the effect of four nebular clusters (footnote to table 1) that might distort the numbers in a few of the squares. The relatively high uniformity of the plates over an area of twenty-five square degrees results in part from the curving of the plates by air pressure throughout the exposures. Cramer Hi-Speed emulsions, specially sensitized, have been used.

Figure 2 presents a similar pair of checkerboard arrays for objects fainter than magnitude 17.0, again for all forty-four plates as well as separately for those in high latitude. This second pair of arrays reflects as much on the chances of discovery and the completeness of the survey over the plates as on possible distance effects on the magnitude estimates. The deviations from uniformity are of the same order of magnitude as the natural uncertainties. The most conspicuous deviations may be the result of unremoved clusterings; but an examination of the mean values for vertical columns and horizontal lines in the different parts of figures 1 and 2 suggests a slight tilt of the photographic plates, during exposure, about a horizontal axis. There is also an indication in these mean values of a "discovery effect," or a slight systematic magnitude correction, for the corner squares. The difference between corners and central areas is best illustrated by the following tabulation of means, which includes all forty-four plates:

	CENTRAL SQUARE	25 CENTRAL SQUARES	9 CENTRAL SQUARES	4 CORNER SQUARES
To magnitude 17.1	250	292	272	310
Magnitude 17.1 and fainter	402	406	402	374
Total range	652	697	675	684

The result for the eighteen high latitude plates is essentially the same as for the forty-four plates. The difference shown for the total range of magnitudes:

$$\text{Central 9 square degrees} - \text{4 corner square degrees} = -9$$

corresponds to an average magnitude gain in the corner square degrees of 0.01, and is therefore negligible. To magnitude 17.1, the difference center *minus* corners of fourteen per cent corresponds to a gain of $0^m.09$, and for the objects fainter than $17^m.0$ there is a loss of $0^m.04$.

Obviously the checkerboard arrays provide a very sensitive means for plate analysis, even when galaxy counts rather than star counts are used. In view of the relative smallness of the derived magnitude deviations, compared with the natural irregularities and the possible uncertainties of the magnitude system, no corrections have been applied to the magnitudes on account of the position of the objects on the plates.

It has seemed advisable, however, to reduce the magnitude estimates by

Miss Jones on the *MC* plates (16-inch Metcalf refractor) to the system of the *A* plates (24-inch Bruce telescope), which cover a much larger part of the sky. The reduction to the Bruce system was carried out before the foregoing examination of the magnitudes was begun; it was simplified by a study of long exposures made by both telescopes on the same rich high latitude fields. Plate *MC* 27639 was measured and the results published by C. K. Seyfert.[3] The magnitudes given by him have been reduced to the Bruce system through the intermediary of measures made by Miss Frances Wright on the same plate.

It will be seen from an inspection of the cumulative totals for each plate, at each tenth of a magnitude from 15.4 to 17.8 (table 2), that the limit of completeness is almost uniformly at 17.5. The limits of the stellar se-

					MEAN
378	461	484	489	407	444
364	418	403	432	396	403
337	362	402	394	377	374
415	401	370	440	380	401
374	455	441	421	339	406
374	419	420	435	380	406

MEAN

					MEAN
323	388	416	430	351	382
322	365	338	382	341	350
275	304	349	346	331	321
340	323	289	377	326	331
306	379	374	374	288	344
313	352	353	382	327	346

FIGURE 2

Total numbers of galaxies of magnitude 17.1 and fainter in each square degree for 44 plates (left), and for 18 plates of highest latitude.

quences (faintest stars clearly detectable) are much fainter, and in the mean we have, from the data in table 1,

$$m_s - m_n = 0.67 \pm 0.03 \text{ (m. e.)}.$$

For many of the plates in low latitude the population of faint galaxies is too small for dependable estimation of the magnitude to which the survey appears to be complete.

4. Summary.—(*a*) The present paper contains in summarized form information on the apparent photographic magnitudes of some twenty-two thousand external galaxies lying in a declination belt five degrees wide, centered at $+43^{\circ}.6$.

(*b*) A thousand of the objects are brighter than or equal to magnitude 15.5. The average plate limit for stars is magnitude 18.12; the nebular survey is complete only to magnitude 17.5, but 4400 of the measured galax-

ies within the twenty-five square degrees area are fainter than that magnitude.

(*c*) The distribution over the plates has been used to examine the nature of the various systematic errors of discovery and measurement. An error arising from plate tilt is intimated; but there is very little suggestion of error from neglected distance correction (figures 1 and 2).

(*d*) The belt at $+43^{\circ}.6$ crosses the Milky Way twice, and the results obtained are therefore useful in examining the effect, on the distribution of external galaxies, of interstellar space absorption along the Milky Way. Qualitatively the absorption is clearly shown in the last three columns of table 1. This latitude effect and the magnitude-frequency curves will be more closely studied in a following paper.

[1] These PROCEEDINGS, **26**, 166–176 (1940); *Harvard Reprint* 194.

[2] Seares, Kapteyn and van Rhijn, *Mt. Wilson Obs. Papers*, Vol. IV (1930).

[3] *Harv. Ann.*, **105**, No. 10, 226 (1937).

INHIBITION OF CARBOXYLASE BY THIAZOLE PYROPHOSPHATE

BY EDWIN R. BUCHMAN, ERIK HEEGAARD AND JAMES BONNER

GATES AND CRELLIN LABORATORIES OF CHEMISTRY AND WILLIAM G. KERCKHOFF LABORATORIES OF THE BIOLOGICAL SCIENCES, CALIFORNIA INSTITUTE OF TECHNOLOGY

Communicated August 9, 1940

In the course of investigations dealing with vitamin B_1 (thiamin) analogs, and having as their goal the furtherance of our knowledge of the physiological rôle of the vitamin, we have observed that a number of substances having a structure similar to that of cocarboxylase (thiamin pyrophosphate[1]), although not themselves able to replace the latter in the enzymatic decarboxylation of pyruvic acid, are able to inhibit markedly the activity of the carboxylase system. The most active inhibitant studied is the pyrophosphoric acid ester of the thiamin thiazole moiety (the thiazole pyrophosphate portion of the cocarboxylase molecule), which will be referred to as "thiazole pyrophosphate."

The experiments were carried out with the usual Warburg manometric technique. Each vessel contained 0.2 g. dried brewers' yeast which had been washed with alkaline phosphate buffer to remove cocarboxylase. The yeast was suspended in 2.0 ml. 0.2 *M* phosphate (pH 6.2) to which was added 0.5 ml. of 0.1 *M* sodium pyruvate solution (pH 6.2) containing 0.1 mg. Mg^{++} and 0.1 mg. Mn^{++}. In the side arm of each vessel was placed 1.0 ml. aqueous solution containing 4 γ cocarboxylase (synthetic[2])

and varying amounts of thiazole pyrophosphate; this solution was then tipped into the main part of the vessel after temperature equilibrium had been reached. Measurements were made at 25°C.

The thiazole was pyrophosphorylated by the method[3] which Weijlard and Tauber used in the case of thiamin. The crude material was purified by conversion into the silver salt[4] and from this we obtained a crystalline manganese salt which was used in the experiments tabulated below:

TABLE 1

INHIBITION OF CARBOXYLASE BY INCREASING AMOUNTS OF THIAZOLE PYROPHOSPHATE (MANGANESE SALT)

VESSEL NO.	1	2	3	4	5	6
Thiazole pyrophosphate (Mn salt)	0	4 γ	8 γ	16 γ	32 γ	80 γ
Cmm. CO_2 in 30 min.	412	388	385	339	226	41

Free thiazole pyrophosphate, liberated in non-crystalline form from the silver salt, gave results consistent with the above. The inhibition is not due to a change in pH; moreover, pyruvic acid decarboxylation is not affected by free thiazole, thiazole monophosphate or sodium pyrophosphate.

We believe that the observed inhibition phenomenon is to be explained on the basis of a competition between cocarboxylase and thiazole pyrophosphate for the specific carboxylase protein with which the two are similarly able to combine. The introduction, together with cocarboxylase, of a substance also capable of combining with the specific protein but giving an inactive "enzyme analog" results in a lowered rate of pyruvate decarboxylation. This interpretation receives support from experiments in which the thiazole pyrophosphate was not added to the protein simultaneously with the cocarboxylase but at a definite time interval before the addition of the latter. It was found that under these conditions the rate of CO_2 production was initially lower than when coenzyme and inhibitor were added together.

Since it is logical to assume that the protein-inhibitor bond is similar to that between protein and cocarboxylase, this latter must therefore be joined to the protein through the pyrophosphate group,[5] which is common to both it and the inhibitor. Thus we have independent confirmation of the prevailing view[6] regarding the nature of this binding, which view was until now supported almost entirely by analogy with the alloxazine-proteid complex.[7]

We conclude that there has been demonstrated here a not hitherto recognized type of competitive inhibition of enzyme reactions, caused by competition not between substrate and inhibitor but between coenzyme and inhibitor. The importance of this type of inhibition must be emphasized because its study may throw light on structural relationships in protein chemistry and on enzyme reaction kinetics and also because there is the

possibility that such inhibition mechanisms are of significance in the chemistry of the cell.

The authors wish to express their gratitude to the Research Corporation for financial aid and to Dr. R. T. Major of Merck and Co., Inc., for generous gifts of cocarboxylase and other chemicals.

[1] Lohmann, K., and Schuster, P., *Biochem. Z.*, **294**, 188 (1937).

[2] Weijlard, J., and Tauber, H., *Jour. Amer. Chem. Soc.*, **60**, 2263 (1938).

[3] We are indebted to Mr. J. Weijlard (Merck and Co., Inc.) who kindly communicated to us the results of unpublished experiments on the pyrophosphorylation of thiazole.

[4] Lohmann and Schuster (ref. 1, page 196) have obtained the silver salt of thiazole pyrophosphate after cleavage of cocarboxylase by sulfite.

[5] A binding through the agency of some other grouping in the cocarboxylase molecule is also to be expected.

[6] See Stern, K. G., and Melnick, J. L., *Jour. Biol. Chem.*, **131**, 610 (1939); Bersin, T., *Kurzes Lehrbuch der Enzymologie*, page 96, Akademische Verlagsgesellschaft, 1938.

[7] Kuhn, R., and Rudy, H., *Ber. Deut. Chem. Ges.*, **69**, 2563 (1936).

ON HOMOTOPY GROUPS

BY SAMUEL EILENBERG

DEPARTMENT OF MATHEMATICS, UNIVERSITY OF MICHIGAN

Communicated July 25, 1940

1. We shall denote by K a finite polyhedron with a fixed cell-decomposition and an orientation attached to each cell. K^m will stand for the subpolyhedron consisting of all cells of K of dimension $\leqslant m$. By definition $K^{-1} = 0$.

Let G be an abelian group and Y a topological space. A triple (K, A^n, f), where A^n is an n-chain in K with coefficients in G and f is a continuous mapping $f(K) \subset Y$, is called a *continuous* n-chain in Y with coefficients in G. Such n-chains and $(n+1)$-chains may serve to define a homology group $\mathfrak{H}^n(Y, G)$.[1]

2. Let y_0 be a fixed point of Y. An n-chain (K, A^n, f) will be called an (n, m)-chain if $f(K^{m-1}) = y_0$ where $0 \leqslant m \leqslant n$. Using the (n, m)-chains and the $(n+1, m)$-chains we can define a new homology group $\mathfrak{H}^{n, m}(Y, G)$.

In the definition of $\mathfrak{H}^{n, m}$ it is essential to specify the point y_0. However, if y_1 is any point which may be joined by an arc to y_0, then the resulting group is isomorphic with the initial group.

3. Let $\pi_i(Y)$ be the ith homotopy group[2] of Y with the point y_0 as origin $(i > 0)$. It is convenient to introduce the notation $\pi_0(Y) = 0$ to indicate that Y is arcwise connected. It is well known that if $\pi_i(Y) = 0$ for $i < m$

then every mapping $f(K) \subset Y$ is homotopic to a mapping $f_1(K) \subset Y$ such that $f_1(K^{m-1}) = y_0$. In particular, it follows that every continuous r-chain in Y is "homotopic" to an (r, m)-chain in Y. Hence

THEOREM I. *If $\pi_i(Y) = 0$ for $i < m$ then $\mathfrak{H}_i^{n, m}(Y, G)$ and $\mathfrak{H}_i^n(Y, G)$ are isomorphic.*

As particular cases we obtain:

(1) *$\mathfrak{H}_i^{n, 0}(Y, G)$ and $\mathfrak{H}_i^n(Y, G)$ are isomorphic.*

(2) *If Y is arcwise connected then $\mathfrak{H}_i^{n, 1}(Y, G)$ and $\mathfrak{H}_i^n(Y, G)$ are isomorphic.*

(3) *If $\pi_i(Y) = 0$ for $i < n$ then $\mathfrak{H}_i^{n, n}(Y, G)$ and $\mathfrak{H}_i^n(Y, G)$ are isomorphic.*

4. Take, now, the integers T as the coefficient group. Let an (n, n)-chain $A^n = (K, A^n, f)$ be given where

$$A^n = \sum_i \alpha_i \sigma_i^n \quad \alpha_i \text{ integers.}$$

Since $f(K^{n-1}) = y_0$ the boundary of the oriented n-cell σ_i^n is mapped into y_0, the mapping $f(\sigma_i^n)$ determining therefore uniquely an element $d(f, \sigma_i^n)$ of $\pi_n(Y)$. Set

$$d(A^n) = \sum_i \alpha_i d(f, \sigma_i^n).$$

For $n > 1$, $d(A^n)$ is an element of $\pi_n(Y)$, for $n = 1$ the fundamental group $\pi_1(Y)$ may not be abelian and $d(A^1)$ is defined only modulo the commutator subgroup of $\pi_1(Y)$. If A^n is the boundary of an $(n + 1)$-cell of K then it is clear that $d(A^n) = 0$. Hence $d(A^n) = 0$ if A^n bounds in K, and d is a homomorphic mapping of $\mathfrak{H}_i^{n, n}(Y, T)$.

THEOREM II. *For $n > 1$ the operation d establishes an isomorphism of $\mathfrak{H}_i^{n, n}(Y, T)$ and $\pi_n(Y)$.*

For $n = 1$ the operation d establishes an isomorphism of $\mathfrak{H}_i^{1, 1}(Y, T)$ and the factor group of $\pi_1(Y)$ by its commutator subgroup.

Using (2) and (3) we deduce from Theorem II the following two known results:

(4) *If Y is arcwise connected then $\mathfrak{H}_i^1(Y, T)$ is isomorphic to the factor group of $\pi_1(Y)$ by its commutator subgroup.*

(5) *If $\pi_i(Y) = 0$ for $i < n (n > 1)$ then $\mathfrak{H}_i^n(Y, T)$ and $\pi_n(Y)$ are isomorphic.*[3]

5. In view of Theorem II we may denote for $n > 1$ the group $\mathfrak{H}_i^{n, n}(Y, G)$ by $\pi_n(Y, G)$ and call it the nth homotopy group of Y with coefficients from G. These groups do not furnish any essentially new invariants of Y as they can be computed using $\pi_n(Y)$ and G.[4]

6. The groups $\mathfrak{H}_i^{n, m}$ can also be defined relative to a subset Y' of Y which contains y_0.

[1] The mechanism defining this group is quite analogous to the one used by N. E. Steenrod, "Regular Cycles of Compact Metric Spaces," *Ann. Math.*, **41** (1940), in print.

[2] W. Hurewicz, *Proc. Akad. Amsterdam*, **38**, 112–119 (1935).

[3] W. Hurewicz, *Ibid.*, **38**, 521–528 (1935).

[4] For other generalizations of the homotopy groups see N. Aronszajn, *C. R.* (*Paris*), **202**, 1475–1478 and 1643–1645 (1936).

ON A GENERAL RULE CONCERNING THE HALF-VALUE PERIODS OF ELECTRON-EMITTING NUCLEI

BY ARTHUR E. HAAS

DEPARTMENT OF PHYSICS, UNIVERSITY OF NOTRE DAME

Communicated August 8, 1940

In an earlier paper[1] the author has pointed out some periodicities in the properties of stable nuclei. In the following it will be shown that some periodicities also reveal themselves if we consider the half-value periods of unstable electron (β^-)-emitting nuclei.

A table which was recently published by Livingood and Seaborg[2] contains more than 300 unstable nuclei. Among them are 93 which are classified as emitting β^--particles (without positrons) and as belonging either to class *A*, i.e., isotope and element certain, or at least to class *B*, i.e., element certain, isotope probable. To these 93 nuclei are to be added about seven more, the discovery of which has been published only since January, 1940.[3]

We may now arrange these unstable β^--emitters in series according to their "isotopic number." By the latter the difference $N - P$ or, what amounts to the same, $A - 2Z$ is understood where N is the number of neutrons, P of protons, A the mass number and Z the charge number. We shall, however, distinguish between such series for which the charge number is even and such for which the charge number is odd. Table 1 contains all sequences of two or more consecutive β^--emitters of classes *A* and *B*; only those β^--emitters are omitted for which we know neither a preceding nor a succeeding nucleus ($_{Z-2}X^{A-4}$ or $_{Z+2}X^{A+4}$) belonging to class *A* or *B*.

From table 1 we may formulate the general rule that in each sequence the half-value period increases from place to place. Although table 1 contains some 60 nuclei, only four nuclei do not fit into the scheme of this rule, namely $_{11}Na^{24}$ (I = 2), $_{21}Sc^{48}$ (I = 6), $_{28}Ni^{63}$ (I = 7), perhaps $_{49}In^{112}$ (I = 14), and finally in the high sequences I = 22 and 36 the nucleus with longer life precedes that with shorter life, in contradiction to the rule.

We find as another rule that those β^--emitters for which both indices, i.e., charge and mass numbers, are odd, would be transformed into stable

nuclei by addition of a proton. The only possible exception to this rule is the β^--emitter $_{55}Cs^{139}$ since the barium isotope $_{56}Ba^{140}$ is not known, although it might be suspected for other reasons.[4]

TABLE 1

SEQUENCES OF β^--EMITTERS WITH THE SAME ISOTOPIC NUMBER

I = 2 (odd)	$_5B^{12}(A)$ 0.022 s	$_7N^{16}(A)$ 8 s	$_9F^{20}(A)$ 12 s	$_{11}Na^{24}(A)$ 14.8 h	$_{13}Al^{28}(A)$ 2.4 m	$_{15}P^{32}(A)$ 14.30 d	$(_{17}Cl^{36}(A)$ >1 y$)$
I = 2 (even)	$_2He^6(A)$ 0.8 s	$_4Be^{10}(A)$ 380 y	$_6C^{14}(A)$ very large				
I = 3	$_8O^{19}(A)$ 31 s	$_{10}Ne^{23}(A)$ 40 s	$_{12}Mg^{27}(A)$ 10.2 m	$_{14}Si^{31}(A)$ 170 m	$_{16}S^{35}(A)$ 88 d		
I = 4	$_{17}Cl^{38}(B)$ 37 m	$_{19}K^{42}(A)$ 12.4 h	$_{21}Sc^{46}(A)$ 85 d				
I = 5	$_{18}Ar^{41}(A)$ 110 m	$_{20}Ca^{45}(A)$ 180 d					
I = 6	$_{21}Sc^{48}(A)$ 44 h	$_{23}V^{52}(A)$ 3.9 m	$_{25}Mn^{56}(A)$ 2.59 h	$_{27}Co^{60}(A)$ 7 y			
I = 7	$_{22}Ti^{51}(A)$ 2.9 m	$_{24}Cr^{55}(A)$ 1.6 h	$_{26}Fe^{59}(A)$ 47 d	$_{28}Ni^{63}(A)$ 2.6 h			
I = 8	$_{29}Cu^{66}(A)$ 5 m	$_{31}Ga^{70}(A)$ 20 m	$(_{33}As^{74}(A)$ 17 d$)$				
I = 12	$_{33}As^{78}(A)$ 65 m	$_{35}Br^{82}(A)$ 34 h	$_{37}Rb^{86}$? ?	$_{39}Y^{90}(A$ 60 h			
I = 13	$_{35}Br^{83}(A)$ 65 m	$_{37}Rb^{87}(A)$ very large					
I = 14	$_{45}Rh^{104}(A)$ 44 s	$_{47}Ag^{108}(A)$ 2.3 m	$_{49}In^{112}(B)$ 72 s or 2.7 d				
I = 15	$_{43}Ms^{101}(A)$ 9 m	$_{45}Rh^{105}(B)$ 46 d					
I = 16	$_{47}Ag^{110}(A)$ 22 s	$_{49}In^{114}(B)$ 48 d					
I = 17	$_{43}Mo^{101}(B)$ 24 m	$_{44}Ru^{105}(B)$ 20 h					
I = 19	$_{46}Pd^{111}(A)$ 17 m	$_{48}Cd^{115}(A)$ 2.5 d					
I = 20	$_{51}Sb^{128}(A)$ 2.8 d	$_{53}I^{128}(A)$ 13.0 d					
I = 25 (even)	$_{50}Sn^{123}(B)$ 9 m	$_{52}Te^{129}(A)$ 70 m					
I = 25 (odd)	$_{51}Sb^{127}(A)$ 80 h	$_{53}I^{131}(A)$ 8.0 d					
I = 27	$_{52}Te^{131}(A)$ 25 m	....	$_{56}Ba^{139}(A)$ 86 m				
I = 32	$_{67}Ho^{166}(B)$ 35 h	$_{69}Tm^{170}(A)$ 105 d					

It might be noteworthy that some regularity reveals itself also in the case of the series I = minus 1, which consists of positron-emitters. As is seen from table 2, the half-value periods decrease monotonically with the charge number up to Si (Na and Al being missing), irrespective whether the charge number is even or odd.

TABLE 2

THE SERIES I = −1, CONTAINING π-EMITTERS

$_6C^{11}(A)$	$_7N^{13}(A)$	$_8O^{15}(A)$	$_9F^{17}(A)$	$_{10}Ne^{19}(A)$	$_{12}Mg^{23}(A)$	$_{14}Si^{27}(A)$
21.0 m	9.93 m	126 s	70 s	20.3 s	11.6 s	3.7 s

[1] Haas, A. E., *Proc. Nat. Acad. Sci.*, **26**, 305 (1940).

[2] Livingood, J. J. and Seaborg, G. T., *Rev. Mod. Phys.*, **12**, 30 (1940).

[3] Walke, H., *Phys. Rev.*, **57**, 163 (1940); **57**, 177 (1940); Pollard, E., *Ibid.*, **57**, 241 (1940); Seaborg, G. T., Livingood, J. J., and Kennedy, J. W., *Ibid.*, **57**, 363 (1940); Ruben, S. and Kamen, M. D., *Ibid.*, **57**, 549 (1940); Sagane, R., *et al.*, *Ibid.*, **57**, 750 (1940); Amaki, T., *et al.*, *Ibid.*, **57**, 751 (1940); Kennedy, J. W. and Seaborg, G. T., *Ibid.*, **57**, 843 (1940).

[4] Haas, A. E., loc. cit.

THE EFFECT OF FAST NEUTRONS ON THE CHROMOSOMES OF TRADESCANTIA

BY NORMAN GILES

BIOLOGICAL LABORATORIES, HARVARD UNIVERSITY

Communicated August 8, 1940

The development of the cyclotron as a powerful and convenient source of high energy neutrons has made available another type of radiation for use in biological studies. Already several investigations have indicated certain differential effects of neutrons as compared with x-rays and gamma rays on biological materials. In several cases a greater relative effectiveness has been found for a given total amount of ionization produced by the recoil protons resulting from neutron bombardment as compared with an equal amount of ionization produced by other types of radiation, particularly secondary electrons resulting from x-radiation and gamma rays (Lawrence and Lawrence,[1] Zirkle and Aebersold,[2] Zirkle, Aebersold and Dempster,[3] Zirkle and Lampe,[4] Gray and Read,[5] Spear, Gray and Read[6]). In most of these studies rather general physiological responses have been observed. Of particular interest to the cytogeneticist is the effect of radiation on genes and chromosomes. The production of mutations has been demonstrated in Habrobracon by Whiting,[7] in Drosophila by Nagai and Locher[8] and Timoféeff-Ressovsky and Zimmer,[9] and in mice by Snell.[10] Chromosome aberrations have been produced by Marshak[11,12] in root tips of *Vicia faba*, *Lycopersicon, esculentum*, and *Pisum sativum* and by Nishina, Sinotô and Sâtô[13] in root tips of *Vicia faba*. In the present study some preliminary observations of the effect of fast neutrons on the microspore chromosomes of Tradescantia are reported.

Methods.—The inflorescences from a clone of a dipoloid Tradescantia which has been used in extensive x-ray experiments by Sax[14] were also used in the present investigation. The flowering stalks, kept in cardboard containers, were placed in the beam of neutrons produced by bombarding a beryllium target with 11 Mev deuterons in the Harvard cyclotron. No shielding to exclude radiations other than neutrons was attempted in these preliminary experiments, but certain control experiments which will be discussed later were carried out. The intensity of the radiation was varied by varying the distance of the inflorescences from the target and keeping the time of exposure constant. The ionization produced by the neutrons was measured for each exposure, and readings are given in "n" units, an "n" unit being defined as that amount of ionization produced by neutrons which gives the same reading on a bakelite Victoreen thimble ionization chamber as does one roentgen of x-rays. Following treatment, the flowering stalks were kept in the greenhouse. Observations were made at the first post-meiotic division in the microspore at suitable periods following treatment by the use of aceto-carmine smear preparations.

Experimental Results.—The effects of neutrons on chromosomes are qualitatively the same as those produced by x-rays. Observations were made at twenty-four to thirty hours and at five days following neutron treatment. At the earlier stage only chromatid aberrations were observed, and at the later, only chromosome aberrations. The configurations were the same as those resulting from chromosome breakage and fusion of broken ends following x-ray treatment as described and diagrammed by Sax.[14]

Quantitatively, however, the results obtained with neutrons differed in two major respects from those obtained by irradiation of developing Tradescantia microspores with x-rays. In the first place, a neutron dose of a given intensity in "n" units was much more effective in producing aberrations than an equal dose of x-rays in "r" units. The data on which the comparisons are based are presented in table 1.

Chromatid dicentrics, resulting from breakage of the two sister chromatids of a single chromosome followed by fusion of broken ends to produce a dicentric and an acentric fragment, were used as these have been found to show an approximately linear proportionality to dosage in x-ray experiments and to be independent of the time factor. In the neutron experiments also the percentage of breaks has been found to be approximately proportional to dosage (Fig. 1). However, for equal doses of "n" and "r" units, neutrons are about 16 or 17 times as effective in producing aberrations as x-rays.

The second difference between the neutron and the x-ray results was in the shape of the dosage curves for exchange break aberrations. In the case of x-rays it has been shown that both chromatid and chromosome

exchanges increase as the square of the dosage. With neutron treatment, however, it seems clear that both types of exchange aberrations are approximately proportional to dosage. The data for chromatid exchange breaks are presented in table 2.

TABLE 1

THE RELATION BETWEEN NEUTRON DOSAGE AND CHROMATID DICENTRICS; TIME CONSTANT. THE RATIO OF EFFECTIVENESS IN THE PRODUCTION OF CHROMATID DICENTRICS OF EQUAL TOTAL IONIZATION BY NEUTRONS AND X-RAYS. EXAMINED AT 30 HOURS

DOSE IN "n" UNITS	TOTAL[1] CHROMOSOMES	CHROMATID DICENTRICS	% B[2]	% B PRODUCED BY EQUAL X-RAY DOSE[3]	RATIO: NEUTRONS / X-RAYS
10.1	2232	85	3.8	0.24	15.8
14.6	2391	155	6.3	0.37	17.0
21.0	2954	265	9.1	0.55	16.5

[1] Figures for total chromosomes which are not multiples of six in this and the following tables indicate occasional microspores with seven chromosomes as a result of nondisjunction of meiosis.

[2] The values for % B (= per cent breaks) given in this and the following tables are averages for several slides and consequently cannot be calculated directly from the figures cited in the tables.

[3] Calculated from the equation % $B = (D/45)^{1.1}$ (Sax[14]).

The data for chromosome exchange breaks, including both rings and dicentrics, represent the combined results of two separate experiments, both of which gave similar results. The figures are given in table 3 and the relations between percentage of breaks and dosage for the different break types are plotted in figure 1.

TABLE 2

THE RELATION BETWEEN NEUTRON DOSAGE AND CHROMATID EXCHANGE BREAKS; TIME CONSTANT. EXAMINED AT 30 HOURS

DOSE IN "n" UNITS	TOTAL CHROMOSOMES	EXCHANGES	RINGS	TOTAL	% B*
10.1	2232	32	4	36	3.2
14.6	2391	50	8	58	4.9
21.0	2954	98	14	112	8.0

* Each aberration involves two breaks.

In order to exclude the possibility that slow neutrons were producing the effects observed, a control experiment was carried out. Two similar sets of inflorescences were used, but one was completely surrounded by sheet cadmium 0.106 cm. thick to filter out the slow neutrons. The two sets were placed at equal distances from the target and irradiated at the same time. The results are shown in table 4.

TABLE 3

THE RELATION BETWEEN NEUTRON DOSAGE AND CHROMOSOME ABERRATIONS; TIME CONSTANT. EXAMINED AT 5 DAYS

DOSE IN "n" UNITS	TOTAL CHROMOSOMES	DICENTRICS	RINGS	TOTAL	% B*
10.1	3384	32	12	44	2.6
13.5	3617	62	26	88	4.9
14.6	3497	57	20	77	4.4
21.0	3678	76	22	98	5.3
27.0	3604	86	36	122	6.8
32.3	4783	163	42	205	8.6
51.0	4313	200	66	266	12.5

* Each aberration involves two breaks.

TABLE 4

TEST OF THE RÔLE OF SLOW NEUTRONS. CADMIUM FILTER USED TO EXCLUDE SLOW NEUTRONS IN *B*. EXAMINED AT 24 HOURS

SERIES	TOTAL CHROMOSOMES	CHROMATID DICENTRICS	%B	EXCHANGES	%B*
A	3350	147	4.45	78	4.6
B (Cd)	3064	143	4.7	62	3.9

* Each aberration involves two breaks.

It can be seen that the percentages of aberrations for the two series are essentially the same, indicating that the fast neutrons rather than the slow ones are producing the effects observed, since a Cd filter of the thickness used removes all thermal neutrons (Livingston and Bethe[15]). In other control experiments no detectable effect on the chromosomes or the division spindles was found when flower buds were exposed to the magnetic field and to the background radiation from the cyclotron chamber.

Technical difficulties prevented any accurate determination of the amount of ionization in the beam due to gamma rays. In the case of the beam produced by the cyclotron of the University of Michigan, Zirkle and Lampe[4] have estimated that the total ionization in tissue due to gamma rays does not exceed twenty per cent. However, there is evidence that the effect of gamma rays on chromosomes is essentially the same as that of x-rays (Muller[16]). Consequently, whatever amount of ionization due to gamma rays is present, it would tend to reduce the magnitude of any differential effect due to neutrons, so that the factor of 16 or 17 is actually a minimum value.

Discussion.—It seems probable that the greater effectiveness of neutrons as compared with x-rays in producing chromosome aberrations must be due to the vastly different nature of the ionization produced in tissue by these two types of radiation. Neutrons produce ionization in tissues principally as a result of collisions with materials containing hydrogen with the resulting ejection of protons. These recoil protons produce a

very intense ionization along their paths as compared with that produced by the secondary electrons generated in matter by x-rays. On the average, one ion pair is formed every 10^{-5} cm. along a β particle track and every 10^{-7} cm. along a proton track. The difference in ion density is thus about 100 times as great for the protons per unit distance of path as for secondary electrons.

There is considerable evidence in the case of x-rays which indicates that the biological effects produced are proportional to the total amount of ionization quite independently of the wave-length of the rays which influences the distribution of the ionization (Packard,[17] Muller,[16] Timoféeff-Ressovsky[18]). However, since the ionization density over the effective x-ray range varies only by a factor of about 10, it is not surprising that there is a greater effect in the case of protons with their much greater ionization density. Investigations producing evidence for this greater effectiveness of recoil protons from neutron bombardment have been cited. Similar results with α particle ionization have been obtained by Zirkle[19] who has shown that the biological effect of this type of radiation on fern spores depends not only on the total amount of ionization but also on the distribution or intensity of this ionization within the cell.

In the present study a given neutron dose in "n" units was found to be from 16 to 17 times as effective in producing chromatid dicentrics as is an equal x-ray dose in "r" units. This particular type of aberration has been found in the x-ray experiments to depend upon "one hit," showing a linear relationship with dosage (Sax[14]). Both threads are broken at the same locus with the subsequent fusion of broken ends to give a dicentric chromatid plus an acentric fragment. In seeking an explanation for the greater effectiveness of neutrons in terms of the difference in ionization density, it is well to keep in mind that all of the chromosome aberrations observed in these experiments result from breakage and subsequent fusion of broken ends in such a way as to produce new and recognizably aberrant configurations. There is good evidence, however, that many breaks are produced by irradiation only to refuse in the original position without resulting in visible aberrations. Consequently, we observe only a fraction, and probably a relatively small one, of the chromosome breaks that actually occur. It seems likely that the greater density of ionization in tissues produced by recoil protons causes a greater number of breaks to occur within certain spatial limits—which are known to play an important rôle in the production of aberrations (Sax[14])—and consequently permits a greater number of aberrant reunions. It seems probable that only occasionally does a single x-ray hit occur in such a way as to break both sister chromatids at prophase; in most cases only one chromatid is broken and such breaks usually refuse. In the case of recoil protons, however, the greater ionization density should result in more cases where a single hit is effective in breaking both

sister chromatids and thus facilitating their subsequent refusion to produce chromatid dicentrics. It is generally assumed in the case of x-radiation that a single ionization is the "hit" which is effective in producing gene mutations and at least some types of chromosome rearrangements. However, the greater effectiveness of proton ionization in producing chromosome aberrations suggests that more than one ionization may be necessary and the differences in the types of curves obtained with neutrons are more easily explained if an electron or proton path is thought of as the effective "hit."

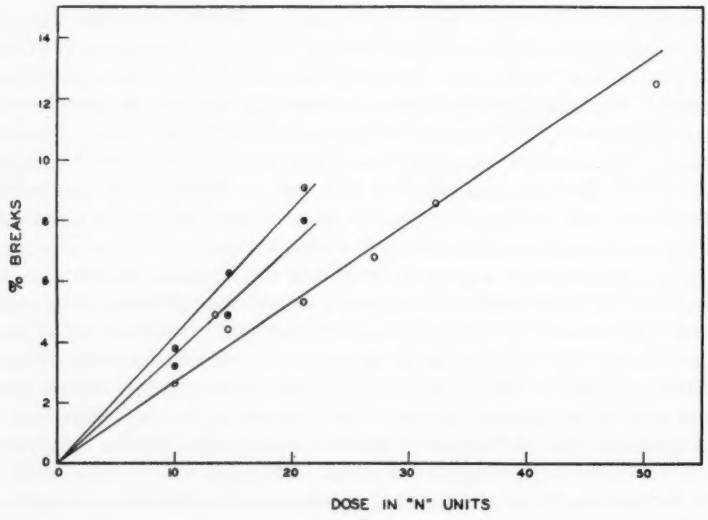

FIGURE 1

Relation between the frequencies of chromatid and chromosome aberrations and neutron dose in "n" units. Time of exposure constant. ◑ = chromatid dicentrics; ● = chromatid exchanges; ○ = chromosome exchanges.

Presumably an explanation based on differences in density of ionization similar to that given for the dicentric chromatids would also explain the greater number of exchange break aberrations with neutron radiation at the dosages studied. However, it is more difficult to compare exchange breaks as to their frequencies with neutron and x-ray treatment since, as indicated earlier, they appear to show different relationships with increasing dosage—an exponential relation for x-rays and a linear relation for neutrons. At low dosages neutrons are considerably more effective in producing aberrations, but as the dosage increases this greater effectiveness becomes less. Actually there should be some dosage above which

x-rays would be more effective than neutrons. In the present experiments this point is not reached within the limits imposed by the possibility of making a correct analysis of the percentage of aberrations in the treated cells. If the curves are extrapolated to higher dosages, however, this point can be determined. In making the calculations it is assumed, as is pointed out below, that the neutron dosage measurements should be multiplied by a factor of 2.5 to give the actual ionization in tissue as compared with x-ray dosages. When this is done for chromosome exchange breaks it is found that above a dose of approximately 500 "n" units, which is taken as equal to 1250 "r" units, x-rays are more effective than neutrons in terms of equal total amounts of ionization. If this general relation holds for all cells it should mean that above some given dosage, the exact value of which would vary with the kind of cell considered, x-rays would be more effective than neutrons in producing cellular injury as measured by chromosome aberrations. If chromosome aberrations play some part in the regression of tumors following irradiation, these results may mean that above certain dosages neutrons would be less efficient in therapy than x-rays.

It is possible that the apparent greater effectiveness of neutrons over x-rays in producing chromosome aberrations may be due to inaccuracies in the methods of comparing the relative amounts of ionization produced by these two types of radiation in tissue. This particular phase of the problem is being investigated in various laboratories. It is pointed out, however, by Marshak[11] that Aebersold and Anslow have unpublished data which indicate that the "n" unit is not larger than the roentgen by a factor greater than 2.5. If this is the case, ionization from recoil protons is still some 6 to 7 times as effective as ionization produced by x-rays in breaking Tradescantia chromosomes.

The experiments of Timoféeff-Ressovsky and Zimmer[9] indicate that neutrons are less effective than x-rays in producing gene mutations in Drosophila. However, since all other work so far has indicated a greater biological effectiveness for neutrons, it seems desirable to await confirmation of the mutation results before attempting a comparison of these with the present findings.

The fact that the exchange break aberrations in the case of the x-ray experiments increase as the square of the dosage when the time factor is kept constant has been taken to indicate that these aberrations are dependent upon two independent x-ray hits (Sax[14]). In the present study these aberration types, particularly the chromosome exchanges, show an approximately linear relationship to dosage. This seems to indicate that both the breaks necessary to produce an aberration are the result of a single hit. The difference in the types of ionization paths in tissue of the electrons and protons may explain this difference, a single proton track being capable of producing two breaks. The relation between dosage and

aberrations is not entirely linear for the chromatid exchange breaks, however, and the presence of an exponent greater than one suggests that two independent hits are sometimes involved. More data are necessary to be certain of the relationship. It is also hoped that the time-intensity and intermittent exposure experiments which are being undertaken will provide critical evidence on this point.

Acknowledgments.—The writer is greatly indebted to Professor K. T. Bainbridge, Doctor B. R. Curtis and Doctor I. A. Getting of the Harvard Physics Department for their coöperation and advice in carrying out these experiments. All the neutron exposures were arranged and measured by Doctor Curtis. He also wishes to express his appreciation to Professor Karl Sax for advice and criticism.

Summary.—The effects of fast neutrons on the microspore chromosomes of Tradescantia have been investigated and compared with the effect of x-rays. Qualitatively the results are the same as those found after x-ray treatment. Quantitatively, however, neutrons appear to differ considerably from x-rays in their effects on chromosomes. For equal total doses in terms of ionization as measured with a bakelite Victoreen "r"-meter neutrons are from 16 to 17 times as effective as x-rays in producing chromatid dicentric aberrations. Also, exchange break aberrations, producing chromatid and chromosome rings and dicentrics, are found to show an approximately linear relationship to dosage instead of the exponential relation found with x-rays. An attempt is made to explain these differences between neutrons and x-rays as a result of the great difference in the types of ionization paths which these two radiations produce in tissue.

[1] Lawrence, J. H., and Lawrence, E. O., *Proc. Nat. Acad. Sci.*, **22**, 124–133 (1936).

[2] Zirkle, R. E., and Aebersold, P. C., *Ibid.*, **22**, 134–138 (1936).

[3] Zirkle, R. E., Aebersold, P. C., and Dempster, E. R., *Amer. Jour. Cancer*, **29**, 556–562 (1937).

[4] Zirkle, R. E., and Lampe, I., *Amer. Jour. Roent. & Rad. Therapy*, **39**, 613–627 (1938).

[5] Gray, L. H., and Read, J., *Nature*, **144**, 439, 479, 509 (1939).

[6] Spear, F. G., Gray, L. H., and Read, J., *Ibid.*, **142**, 1074 (1938).

[7] Whiting, P. W., *Science*, **84**, 68 (1936).

[8] Nagai, M. A., and Locher, G. L., *Genetics*, **23**, 179–189 (1938).

[9] Timoféeff-Ressovsky, N. W., and Zimmer, K. G., *Naturwissenschaften*, **21/22**, 362–365 (1938).

[10] Snell, G. D., *Proc. Nat. Acad. Sci.*, **25**, 11 (1939).

[11] Marshak, A., *Proc. Soc. Exptl. Biol. Med.*, **41**, 176–180 (1939).

[12] Marshak, A., *Proc. Nat. Acad. Sci.*, **25**, 502–510 (1939).

[13] Nishina, Y., Sinotô, Y., and Sâtô, D., *Cytologia*, **10**, 406–421 (1940).

[14] Sax, K., *Genetics*, **25**, 41–68 (1940).

[15] Livingston, M. S., and Bethe, H. A., *Rev. Mod. Physics*, **9**, page 254 (1937).

[16] Muller, H. J., *Jour. Genet.*, **40**, 1–66 (1940).

[17] Packard, C., Chap. XIII in Duggar, B. M., *Biological Effects of Radiation*, McGraw-Hill Co., New York (1936).

[18] Timoféeff-Ressovsky, N. W., *Experimentelle Mutationsforshung in der Vererbungslehre*, pp. 1–181, Theodor Steinkopff, Dresden und Leipzig (1937).
[19] Zirkle, R. E., *Amer. Jour. Cancer*, **23**, 558–567 (1935).

CHROMOSOME HOMOLOGIES IN TWO SUB-SPECIES OF DROSOPHILA VIRILIS

BY HARRISON D. STALKER

DEPARTMENT OF ZOÖLOGY, UNIVERSITY OF ROCHESTER

Communicated July 17, 1940

It has been shown by Hughes[1] that the two sub-species, *Drosophila virilis virilis* Sturtevant, and *Drosophila virilis americana* Spencer (Spencer[2]), differ, not only in their salivary chromosomes, but also with respect to the metaphase chromosome configuration. The *virilis* metaphase plate shows five pairs of rod-shaped chromosomes and one pair of dots. In *americana*, on the other hand, the female metaphase plate consists of one pair of rods, two pairs of V-shaped chromosomes and a pair of dots, while the male has one pair of rods, one pair of V-shaped chromosomes, a pair of dots, and one V-shaped chromosome typically showing somatic pairing with two rod-shaped chromosomes.

Hughes has concluded that one pair of V-shaped chromosomes in *americana*, the pair found in both sexes, corresponds to two rod-shaped chromosomes in *virilis;* also that the other pair of V-shaped chromosomes found in the *americana* female corresponds to an autosome and the *X*-chromosome of *virilis;* while the two rod-shaped chromosomes pairing with the V-shaped chromosome in the *americana* male correspond to a *virilis Y*-chromosome and an autosome. (See Fig. 1.)

The following investigation was undertaken to determine genetically the chromosome homologies existing between the two sub-species. More specifically it was desired to secure genetic confirmation of Hughes's cytological findings in regard to the relationships existing between the *X*, the *Y* and the autosomes in *americana*.

Reciprocal crosses were made between a stock of *virilis* carrying marker genes on all five major chromosomes, and three strains of *americana*. The three *americana* strains were all collected from points within ten miles of Wooster, Ohio. One of them, the "Smithville" strain, is the one which Hughes used in his cytological investigation.

The *virilis* marker stock carried *y* (yellow) on the *X*-chromosome, *va* (varnished) on the second chromosome, *tb* (tiny bristle) on the third chromosome, px^2 (plexus[2]) on the fourth chromosome and *ru* (ruffled) on

the fifth, the sixth, or dot-shaped chromosome being unmarked. The first series of crosses was between *americana* females and *virilis* males.

P_1 *americana* ♀♀ X *y, va, tb, px², ru* (*virilis*) ♂♂

B.C. hybrid ♂♂ X *y, va, tb, px², ru* (*virilis*) ♀♀

Table 1 gives the types and frequencies of the back-cross offspring.

TABLE 1

OFFSPRING FROM BACKCROSS OF HYBRID MALE (*americana* ♀♀ X *virilis* ♂♂), X *virilis* ♀♀

PHENOTYPES	SMITHVILLE		OVERTON		PEE WEE-3	
	♂♂	♀♀	♂♂	♀♀	♂♂	♀♀
y va tb px² ru	96		35		73	
wild		135		53		144
y va tb px²	75		34		88	
ru		108		43		91
va tb ru		121		47		92
y px²	149		59		164	
y px² ru	70		29		42	
va tb		115		50		111

It can be seen that with all three races tested, the back-cross offspring show a complete linkage between *va* and *tb*, thus suggesting that the V-shaped autosomes of *americana* correspond to the second and third chromosomes of *virilis*.

Table 1 shows further that in all cases the females were wild type for

FIGURE 1

A—Metaphase chromosomes of male or female *Drosophila virilis virilis*. *B*—Metaphase chromosomes of female *Drosophila virilis americana*. *C*—Metaphase chromosomes of male *Drosophila virilis americana*.

the characters *yellow* and *plexus²*, while all the males showed these characters. The sex-linked behavior of yellow in the hybrid is in agreement with its location in the *X*-chromosome of *virilis*. The change from an autosomal inheritance of *plexus²* in *virilis* to a sex-linked mode in the hybrid suggests that the *virilis* chromosome carrying *px²*, namely the fourth

chromosome, is joined to the X to form the V-shaped heterochromosome of *americana*.

A reciprocal cross was made to test genetically Hughes's findings of an *americana* Y-chromosome pairing with the X, but not, like the X-chromosome, attached to an autosome.

F_1 *y, va, tb, px², ru (virilis)* ♀♀ X *americana* ♂♂

B.C. hybrid ♂♂ X *y, va, tb, px², ru (virilis)* ♀♀

TABLE 2

OFFSPRING FROM BACKCROSS OF HYBRID MALE (*virilis* ♀♀ X *americana* ♂♂), X *virilis* ♀♀

PHENOTYPES	SMITHVILLE		OVERTON		PEE WEE-3	
	♂♂	♀♀	♂♂	♀♀	♂♂	♀♀
y va tb px² ru	6	2	23	26	54	62
y	1	4	13	15	103	85
y va tb ru	3	5	16	23	56	52
y px²	4	5	16	15	105	58
y va tb px²	2	7	10	23	70	87
y ru	3	5	12	10	13	21
y px² ru	3	1	19	20	22	22
y va tb	2	7	26	10	85	89

In this cross the hybrid male received a Y-chromosome from the *americana* parent. His offspring did not show sex-linked inheritance of any of the *virilis* autosomal genes. Thus we may conclude that there is no preferential segregation exhibited between the Y and any autosome in *americana*, and that the Y is not fused to an autosome, thus corroborating genetically Hughes's cytological findings.

In agreement with the data of table 1, the reciprocal crosses reported in table 2 show again for all three races complete linkage between the genes *va* and *tb*, suggesting a fusion of the second and third chromosomes of *virilis* to form the V-shaped autosome of *americana*.

If the chromosomes that are considered to be homologous on the basis of the marker genes used are so with respect to the majority of their genes, the two sub-species might be schematically represented as follows:

Drosophila virilis virilis

Female:	(*X*)	(IV)	(II)	(III)	(V)	(VI)
	(*X*)	(IV)	(II)	(III)	(V)	(VI)
Male:	(*X*)	(IV)	(II)	(III)	(V)	(VI)
	(*Y*)	(IV)	(II)	(III)	(V)	(VI)

Drosophila virilis americana

Female:	(*X*....IV)	(II....III)	(V) (VI)
	(*X*....IV)	(II....III)	(V) (VI)
Male:	(*X*....IV)	(II....III)	(V) (VI)
	(*Y*) (IV)	(II....III)	(V) (VI)

[1] Hughes, R. D., *Genetics*, **24**, 811 (1939).
[2] Spencer, W. P., *Ibid.*, **23**, 169 (1938).

LINKAGE STUDIES OF THE RAT (RATTUS NORVEGICUS). III

By W. E. Castle and Helen Dean King

University of California and Wistar Institute*

Communicated July 16, 1940

A new mutant character of rats, "hereditary acholuric jaundice" was described in 1938 by C. H. Gunn of the University of Toronto. It was shown that the anomaly results from an overproduction of bile pigment in consequence of an excessive fragility of the erythrocytes. The affected individuals are recognizable by their yellow color at or soon after birth. The yellow pigment also enters the developing hair so that white hair of albino and of piebald individuals is distinctly yellowish in color. Growth is usually retarded in affected individuals and in extreme cases "nervous symptoms are developed such as a wobbly gait or partial paralysis, confined chiefly to the hind limbs." Experimental evidence, according to Gunn, indicates that the lag in growth and the nervous symptoms are associated with an inability of jaundiced rats to use carotene as a source of vitamin A, and that they consequently suffer from a prolonged vitamin A deficiency. He shows that the syndrome is inherited as a simple recessive character, which finding is fully conformed by our own observations.

Through the kindness of Professor John W. MacArthur, in whose laboratory the studies of Gunn were made in part, we received in Berkeley a stock of albino jaundiced rats in September, 1938, planning to make a complete study of the linkage relations of the jaundice gene. Dr. MacArthur had already informed us by letter that he had canvassed the question of possible linkage with the commoner mutant rat genes, agouti, albino and hooded, and found no indication of linkage, as recombination occurred freely after crosses involving each of these genes. This finding also we can confirm. The symbol used by MacArthur for jaundiced is *j*, which usage we shall follow.

The mutant genes for which tests have been made by us for linkage with jaundiced are as follows: (1) *A*, agouti; (2) *c*, albino; (3) *Cu*, curly; (4) Cu_2, curly$_2$; (5) *d*, dilute; (6), *h*, hooded; (7) *hr*, hairless; (8) *k*, kinky; (9) *wo*, wobbly. Other mutant genes for which no tests have been made because they are known to be linked with one or another of the genes already enumerated are the following: (10) *b*, brown and (11) *an*, anemia,

which are linked with *Cu;* (12) *l*, Grüneberg lethal, (13) *p*, pink-eye, (14) *r*, red-eye and (15) *w*, waltzing, all of which are linked with *c*, albino.

It may be stated at once that all linkage tests made by us have given negative results, so that for the present jaundiced must be regarded as the marker of an independent tenth chromosome.

A summary statement of the results of the linkage tests is contained in tables 1 and 2.

Tests were made by a backcross to the double recessive in the case of agouti, albino, curly, curly$_2$, and hooded, the expectation being equality of crossover and non-crossover classes, if no linkage exists. Such equality was found within limits scarcely in any case exceeding the probable error, and so without statistical significance. (See table 1.)

F_2 populations (table 2) were considered conclusive evidence of the non-existence of linkage in the cases of dilution, hairless and kinky, in all of which the critical double recessive recombination class, which could arise only from the union of crossover gametes, was found to be close to or in excess of expectation, whereas if linkage existed this class should be below expectation on the basis of free assortment. Also other classes in these F_2 populations were sufficiently close to expectation to negative the idea of linkage.

In the case of wobbly (table 1) an F_2 test was rendered unreliable and a back-cross test between F_1 and the double recessive was rendered impossible by early death of double recessives, none having attained maturity. Accordingly resort was had to the more laborious but more certain method outlined in a previous paper (Castle, 1939). A cross was made between jaundiced and wobbly. F_1 was crossed to animals carrying neither mutant gene. The resulting young were tested individually for presence of one or the other or both of the mutant genes. These tests were made by mating each individual to be tested to an F_1 animal, which from its pedigree would be known to be a carrier of both mutant genes. If there was no linkage between the two mutant genes, those genes would be expected to recombine freely among the gametes produced by an F_1 individual. Such gametes would then be of four sorts equally numerous, *viz.*, (1) *j* only, (2) *wo* only, (3) both *j* and *wo* and (4) neither *j* nor *wo*. No test mating was rated as conclusive unless it resulted in the production of six or more young. Satisfactory tests were made of 143 animals which fall into four classes as follows:

CARRIERS OF *j* ONLY	CARRIERS OF *wo* ONLY	CARRIERS OF BOTH *j* AND *wo*	CARRIERS OF NEITHER *j* NOR *wo*
32	40	23	48

As regards the contribution of the F_1 parent to the animals tested, carriers of *j* only or of *wo* only would arise from repulsion (non-crossover) gametes of the F_1 parent, whereas carriers of *both* or of *neither* would arise from crossover gametes of the F_1 parent. We thus have information as to the nature

of 173 F_1 gametes. These total 32 + 40 = 72 non-crossover gametes, and 23 + 48 = 71 crossover gametes. The two groups are as nearly equal as possible in an odd number of individuals. They furnish a perfect example of free assortment and show conclusively that no linkage exists between *j* and *wo*.

Conclusion.—The recessive mutant gene jaundiced (*j*) shows linkage with no other known rat gene and thus becomes the marker of a tenth independent chromosome pair.

Acknowledgment is made of valuable assistance rendered by WPA laborers in caring for the experimental animals, project No. 65-1-08-91, unit B-7.

TABLE 1

SUMMARY OF TESTS FOR LINKAGE BETWEEN JAUNDICED (*j*) AND OTHER MUTANT GENES OF THE RAT

BACKCROSSES BETWEEN F_1 AND DOUBLE RECESSIVES	CROSSOVER GAMETES	NON-CROSSOVER GAMETES	DEVIATION AND *P.E.*
j × agouti	87	81	3 ± 4.4
j × albino	59	59	0
j × curly	257	267	5 ± 7.7
j × curly$_2$	72	84	6 ± 4.2
j × hooded	102	113	5.5 ± 4.9
	Test by "lethal method," Castle, 1939		
j × wobbly	71	72	0.5 ± 4.0

TABLE 2

F_2 POPULATIONS. EXPECTED 9:3:3:1, IF NO LINKAGE EXISTS

j × dilution	*JD*	*jd*	*Jd*	*jd*
	89	29	22	17
Expected	*88.2*	*29.4*	*29.4*	*9.8*
j × hairless	*JHr*	*jHr*	*Jhr*	*jhr*
	95	32	22	9
Expected	*89.0*	*29.7*	*29.7*	*9.9*
j × kinky	*JK*	*jK*	*Jk*	*jk*
	352	108	134	51
Expected	*362.7*	*120.9*	*120.9*	*40.3*

*This is the third in a series of reports on coöperative investigations of linkage in the rat. In the first two papers all observations reported were made by King at the Wistar Institute. In this paper the observations were made by Castle in Berkeley.

Castle, W. E., "On a Method for Testing for Linkage between Lethal Genes," *Proc. Nat. Acad. Sci.*, **25**, 593–594 (1939).

Gunn, C. H., "Hereditary Acholuric Jaundice in a New Mutant Strain of Rats," *Jour. Hered.*, **29**, 137–139 (1938).

King, Helen Dean, and Castle, W. E., "Linkage Studies of the Rat (*Rattus norvegicus*)," *Proc. Nat. Acad. Sci.*, **21**, 390–399 (1935).

King, Helen Dean, and Castle, W. E., "Linkage Studies of the Rat (*Rattus norvegicus*). II," *Ibid.*, **23**, 56–60 (1937).

SYNTHESIS OF THIAMIN BY EXCISED ROOTS OF MAIZE*

BY J. EDWARD MCCLARY

DEPARTMENT OF BOTANY, UNIVERSITY OF MISSOURI

Communicated August 13, 1940

Excised roots of many dicotyledonous plants have been cultured indefinitely in a nutrient medium consisting of mineral salts, sugar and one or more accessory growth factors. Some species require the addition of vitamin B_1 (thiamin),[11] others require also vitamin B_6[8] or nicotinic acid or both.[1] According to Robbins[6] the tomato requires only the addition of the thiazole fraction of thiamin but Bonner and Devirian[1] found that thiamin could not be replaced by thiazole for the growth of excised tomato roots.

Similar experiments with monocotyledonous plants have been unsuccessful. Many attempts have been made, particularly with maize (*Zea mays*) and favorable effects have been reported for yeast extract[4, 5] and the extracts of parts of the corn plant or seed,[7] but growth of the tips has not been maintained through more than a few, rarely as many as six, transfers and the rate of growth in the successive transfers has always decreased. Robbins and White[7] and Fiedler[3] found that agar medium was superior to aqueous medium for the growth of excised roots of corn, but a constant growth rate was not maintained through transfers to fresh media and only a few transfers could be made.

Fiedler concluded that it was probably impossible to culture roots of maize indefinitely since it is strictly an annual plant and the growth of the roots is thereby limited. This conclusion is incapable of proof and need not be considered. Robbins and White[7] concluded that organic substances other than glucose as well as minerals not ordinarily supplied were probably essential for the growth of maize roots and the beneficial effect of agar was attributed to impurities that could be assimilated by the roots. Robbins[9] has shown that agar contains traces of growth factors, but the evidence presented herein seems to indicate that the physical properties of the agar solution are, at least in part, responsible for the beneficial effects of the agar.

Evidence is presented in this report that excised roots of maize can be cultured through many transfers without decrease in the growth rate in a solution of mineral salts, glucose and agar, and that under these conditions thiamin is synthesized by the roots.

Materials and Methods.—The varieties of maize used in these experiments were inbred lines and single crosses furnished by the Field Crops Department of the University of Missouri. For the most part the single crosses *RYD* 940 *X* L_3 and *HY X* L_3 were used. The mineral salts used

were Merck's or Baker's C.P. grade, not specially purified. The glucose was cerelose, a pure grade produced by the Corn Products Company. The agar was "Difco, Bacto-agar," granular form. No analysis has been made of the substances and no attempt has been made to purify them. The asparagin was recrystallized four times from hot water. The water was redistilled from Pyrex glass.

The basic mineral solution for the culture of excised root tips was Hoagland's[4] slightly modified, consisting of KNO_3—0.25 g.; $Ca(NO_3)_2.4H_2O$—0.59 g.; $MgSO_4.7H_2O$—0.246 g.; KH_2PO_4—0.068 g.; $Fe_2(SO_4)_3$—0.004 g.; water—1 liter. Due to evidence not necessary to this discussion the mineral solution was modified in the later work by the substitution of KCl for KNO_3 in equivalent parts and the addition of 2 grams per liter of NaCl.

Grains of maize were sterilized in a 0.2% solution of $HgCl_2$ in 50% ethyl alcohol for 1 to 2 minutes. They were washed for at least 10 minutes in distilled water in large test tubes and removed to sterile agar plates without nutrients by means of an aluminum spoon. When the roots were from 1 to 4 cm. in length, tips approximately 2 mm. in length were excised and transferred to the nutrient solution.

Unless otherwise mentioned all cultures of both roots and Phycomyces were grown in 125-ml. Erlenmeyer flasks of Pyrex glass. The flasks were cleaned with chromic acid cleaning solution, rinsed with tap water, distilled water and finally with redistilled water.

For thiamin analyses the Phycomyces method described by Schopfer[10] was used. The medium for the culture of *Phycomyces Blakesleeanus* consisted of $MgSO_4.7H_2O$—0.5 g.; KH_2PO_4—1.5 g.; glucose—100 g.; asparagin—4 g.; water—1 liter. The material to be assayed was ground in a glass mortar and added to 20 ml. of the nutrient medium in 125-ml. flasks. The media were sterilized in an autoclave at 15 lb. pressure for 20 minutes. Each flask was inoculated with 0.2 ml. of a spore suspension made by shaking a tuft of Phycomyces in 50 ml. of distilled water. The cultures were allowed to grow for 10 days at a temperature of 20 to 23 degrees in diffuse light. The mycelia were washed with distilled water and dried at 80 degrees.

Estimation of the quantity of thiamin present was made from controls grown with synthetic thiamin ("Betabion, Merck") at the same time and under the same conditions as the assay cultures.

Experimental Results: Culture of Excised Roots in Aqueous Media.—All attempts to culture excised corn roots through successive transfers to fresh media have failed. The addition of thiamin, nicotinic acid, amino acids, nucleic acid, ascorbic acid and other growth factors have had little or no effect in any combination tried. Aeration of the medium by bubbling air continuously through the solution was without effect. Although, as is shown later, higher concentration of glucose improves agar media for the

growth of corn roots, 2% glucose was optimum in the aqueous media employed.

Culture of Roots in Agar Medium with 2 Per Cent Glucose.—Using Hoagland's solution plus 2% glucose in 1½% agar as the culture medium, the results obtained in this laboratory have been very variable. When the roots were allowed to remain in the original medium for long periods of time, sometimes for 3 or 4 months, they often attained a dry weight of 100 mg. or more, but all attempts to maintain the roots through successive transfers of the tip to fresh media failed. The addition of known growth factors did not improve the solution for the growth of the roots. Extracts of maize seed, leaves or roots of corn, and of yeast were not markedly beneficial even when such extracts were filtered sterile and added to the agar medium at a temperature just high enough to prevent solidification of the agar. It seems probable that the failure of growth under these conditions was not due to the lack of any growth factor that is required in small amounts unless that substance is very unstable or is not absorbed by the roots.

The Culture of Excised Embryos of Maize.—It is probable that when an embryo has a large part of the scutellum attached it will not be deficient in thiamin or other required growth factors until it has made considerable growth. Analysis of the seed for thiamin was made for the purpose of determining how much thiamin could be expected to be present under these conditions, and how rapidly the store of thiamin was depleted by the growing plant. Seed of an F_1 hybrid, *RYD* 940 *X* L_3, were soaked for 24 hours at 15 degrees and germinated on agar plates, without nutrients, under sterile conditions, and in the dark at room temperature. Samples were analyzed for thiamin after 0, 5 and 10 days of germination. The results are shown in table 1.

TABLE 1

ASSAY OF GERMINATING MAIZE SEED FOR THIAMIN

TIME IN DAYS OF GERMINATION	FRACTION OF SEED	THIAMIN, TIMES 10^{-6} MOLE
0	Embryo +	
	Scutellum	0.37
	Endosperm	0.02
	Total	0.39
5	Embryo +	
	Scutellum	0.45
	Endosperm	0.02
	Total	0.47
10	Embryo +	
	Scutellum	0.53
	Endosperm	0.02
	Total	0.55

Although the scutellum and embryo constitute less than 12 per cent of the dry weight of the seed they contain about 94 per cent of the thiamin contained in the seed. There was an increase in the total amount of thiamin in the seed; after 5 days the increase was 21.6 per cent and after 10 days 43.2 per cent of the amount originally contained in the seed.

Embryos with approximately $^1/_4$ of the scutellum attached were excised from germinating seeds and cultured in the dark in Hoagland's solution plus 0.5, 2 and 5 per cent glucose and $1^1/_2$ per cent agar for 21 days. The results are given in table 2.

TABLE 2

THE GROWTH OF THE ROOTS OF CULTURED EMBRYOS WITH ABOUT $^1/_4$ OF THE SCUTELLUM ATTACHED. TIME OF GROWTH WAS 21 DAYS

TISSUE CULTURED	PER CENT OF GLUCOSE	DRY WEIGHT OF ROOTS, MG.
Roots of embryos	0.5	3.0
	2.0	10.8
	5.0	30.6
Excised roots	2.0	9.5

In this experiment glucose was obviously the limiting factor in the lower concentrations. The weight of the roots in the cultures of 5 per cent glucose was more than double the weight of the roots in the lower concentrations. Perhaps more important from the standpoint of this discussion was the fact that in 2 per cent glucose the roots of these plants grew no better than excised roots grown in the same medium. They were similar in appearance, becoming exceedingly thin and stopping growth after about two weeks. In 5 per cent glucose the roots were more nearly normal in appearance and were growing in all cultures at the termination of the experiment after 21 days.

The Culture of Excised Roots of Maize in Agar Medium Plus 5 Per Cent Glucose.—On April 15 excised root tips 2 mm. in length were started in the basic mineral solution plus $1^1/_2$ per cent agar plus 2, 5, $7^1/_2$ and 10 per cent concentrations of glucose. The corn used was a single cross, *RYD* 940 *X* L_3. After 20 days all of the tips in the 2 per cent glucose media were too thin for transferring. Those in $7^1/_2$ and 10 per cent glucose were thick but had grown slowly and were discarded. The roots in media containing 5 per cent glucose were normal in appearance and were growing well. These, 8 in number, were sub-cultured by removing 1 cm. of the tip to fresh media of the same constitution. These roots are still growing well at the time of this writing; they are in the eighth sub-culture and have been growing at an almost constant rate of approximately 8 mm. per day for 115 days. Furthermore, each culture, after the tip was removed, developed branch roots that thickened and grew as well as the main root.

In another experiment, that is being carried on at the time of writing, the tips of the roots have been transferred to fresh media at intervals of three days. They have now been transferred 18 times, the rate of increase in length has not varied appreciably and the tips appear to be perfectly normal and as large in diameter as the original tips. During the last 10 of these transfers the tips have been grown in Pyrex petri dishes (150 × 15 mm.) containing 10 ml. of nutrient medium. Many of these roots have at times become abnormal when they penetrated into the agar medium; but growth was resumed at the original rate when the roots were removed from the agar so that only the tip was in contact with the agar or penetration was very shallow.

Synthesis of Thiamin by Excised Roots of Corn.—Many assays of excised roots that had grown without transfer in the original medium indicated that in every instance the cultured roots contained more thiamin than was present in the original tip and in the nutrient medium. In each transfer of the experiment described above, started April 15, the old roots were allowed to grow, after the tips were removed, until June 14, when the first four cultures were assayed for thiamin. The results are given in table 3.

TABLE 3

ASSAY OF EXCISED ROOTS OF MAIZE FOR THIAMIN

TRANSFER	NUMBER OF OF ROOTS	TIME OF GROWTH, DAYS	AV. DRY WEIGHT OF ROOTS, MG.	AV. AMT. OF THIAMIN PER ROOT $\times 10^{-10}$ MOLE
1	8	60	62.5	1.81
2	8	30	26.5	0.86
3	7	16	16.3	0.54
4	7	9	7.1	0.33
Total thiamin per root through 4 transfers				3.54
Controls, 1 3-mm. tip excised from a germinating grain of corn, average of 15 cultures				0.15
Culture medium for root culture plus asparagin, average per flask				0.02

A single root growing through 4 transfers was found to contain 3.54×10^{-10} moles of thiamin compared with 0.15×10^{-10} in an original root tip and 0.02×10^{-10} moles in the agar medium. The growth of Phycomyces was little better in agar without thiamin than in water without thiamin and evidently only a trace of this substance is to be found in agar. A cultured root contained about 24 times as much thiamin as the original tip and there seems to be no way to account for this increase other than synthesis by the roots from glucose, mineral salts and whatever impurities may be present and available in the medium.

Discussion.—No improvement over results already reported has been made in the culture of excised corn roots in aqueous media. The addition of numerous growth factors and extracts prepared in various ways has failed to improve, materially, the mineral salt plus glucose solution and the growth of the excised roots has consistently been unsatisfactory. The results are similar regardless of the organic substances added to the solution and in view of the results reported with agar cultures it seems doubtful that this unsatisfactory growth is the result of a deficiency of any growth factor. It seems more probable that the physical conditions of the aqueous medium are unsatisfactory for the continued growth of excised maize roots.

The conditions under which sustained growth of excised maize roots can be maintained are yet imperfectly delineated. Growth has been maintained through 18 transfers without appreciable decrease in the rate of elongation of the roots and branch roots have been removed from the latest transfers and in turn maintained in culture. It seems doubtful that any substance in the original tip could still be present in sufficient concentration in the later cultures to control the growth of the roots. However, the fact that root tips often become abnormal and cease to grow, even under the most favorable conditions, if they are allowed to penetrate the agar, and that the growth of such roots will often be resumed if they are placed with only the tip in contact with the agar, indicates that conditions other than nutrient deficiency are responsible for the development of the abnormal condition. The fact that maize plants will grow with their roots in water solution does not necessarily invalidate this argument. When the roots are attached to the aerial part of the plant the vascular system is uninterrupted, nutrients are furnished from the aerial parts and there is not usually glucose in the medium surrounding the roots. This is obviously a system quite different from an excised root growing entirely surrounded by the medium. The most plausible explanation of the effect of removing a root from the agar is that aeration is better; and yet it is difficult to understand how a system could be deficient in air when air bubbles are continuously passing through the system, or how a layer of medium only 2 mm. in thickness could be deficient in air. Although it seems likely that physical conditions of the medium are important, the nature of the factors involved are unknown.

The evidence for the synthesis of thiamin by excised roots of maize seems to be conclusive. Whether glucose and mineral salts alone are sufficient for the synthesis of thiamin and for continued growth of the roots is still open to some doubt. It is possible that traces of impurities in the agar and glucose can be utilized. Whatever impurities are present are not effective in improving materially the growth of excised roots in aqueous medium and it is doubtful if their presence is of great importance in the agar

media. It is clear that excised roots of corn can utilize materials that are not effective in replacing thiamin for the growth of Phycomyces and convert them into thiamin or its equivalent as measured by the growth of Phycomyces.

Summary.—1. Efforts to improve an aqueous solution of mineral salts and glucose for the growth of excised maize roots by the addition of known growth factors or of various extracts were not successful. Aeration did not improve the solution for the growth of the roots.

2. Excised maize roots were maintained in a medium consisting of mineral salts, 5% glucose and $1^1/_2$ per cent agar for 115 days through 18 transfers without appreciable decrease in the rate of elongation of the roots.

3. Thiamin was synthesized in significant quantities by excised roots of maize growing in the mineral salt, glucose and agar solution.

4. Evidence is presented that the physical conditions of the media were probably limiting factors for the growth of maize roots in an aqueous medium and under certain conditions in agar medium.

* A portion of a dissertation to be presented to the Graduate Faculty of the University of Missouri in partial fulfilment of the requirements for the degree of Doctor of Philosophy.

[1] Bonner, James, and Devirian, Philip S., "Growth Factor Requirements of Four Species of Isolated Roots," *Am. Jour. Bot.*, **26**, 661–665 (1939).

[2] Bonner, James, and Erickson, J., "The Phycomyces Assay for Thiamin (Vitamin B_1): the Method and Its Chemical Specificity," *Ibid.*, **25**, 685–692 (1938).

[3] Fiedler, Herbert, "Entwicklungs und Reizphysiologische Untersuchungen an Kulturen isolierter Wurzelspitzen," *Zeit. f. Bot.*, **30**, 385–436 (1937).

[4] Hoagland, D. R., and Broyer, T. C., "General Nature of the Process of Salt Accumulation by Roots with Description of Experimental Methods," *Pl. Physiol.*, **11**, 477–507 (1936).

[5] Robbins, William J., "Effect of Autolyzed Yeast and Peptone on the Growth of Excised Corn Root Tips in the Dark," *Bot. Gaz.*, **74**, 59–79 (1922).

[6] Robbins, W. J., and Bartley, Mary A., "Thiazole and the Growth of Excised Tomato Roots," *Proc. Nat. Acad. Sci.*, **23**, 385–388 (1937).

[7] Robbins, W. J., and White, Virginia B., "Effect of Extracts from the Corn Plant on the Growth of Excised Root Tips," *Bot. Gaz.*, **98**, 520–534 (1937).

[8] Robbins, W. J., and Bartley, Mary A., "Vitamin B_6, a Growth Substance for Excised Roots," *Proc. Nat. Acad. Sci.*, **25**, 1–3 (1939).

[9] Robbins, W. J., "Growth Substances in Agar," *Am. Jour. Bot.*, **26**, 772–778 (1939).

[10] Schopfer, W., and Jung A., "Un test vegetal pour l'aneurine. Méthode, Critique, et Résultats," *Compt. Rend. V Congres Int. Tech. et Chim. des Ind. Agr. Schevingen*, **1**, 22–34 (1937).

[11] White, P. R., "Vitamin B_1 in the Nutrition of Excised Tomato Roots," *Pl. Physiol.*, **12**, 803–811 (1937).

[12] White, P. R., "Cultivation of Excised Roots of Dicotyledonous Plants," *Am. Jour. Bot.*, **25**, 348–356 (1938).